BEST AVAILABLE IMAGES

Defective images within this document are accurate representations of the original documents submitted by the applicant.

Defects in the images may include (but are not limited to):

- BLACK BORDERS
- TEXT CUT OFF AT TOP, BOTTOM OR SIDES
- FADED TEXT
- ILLEGIBLE TEXT
- SKEWED/SLANTED IMAGES
- COLORED PHOTOS
- BLACK OR VERY BLACK AND WHITE DARK PHOTOS
- GRAY SCALE DOCUMENTS

IMAGES ARE BEST AVAILABLE COPY.

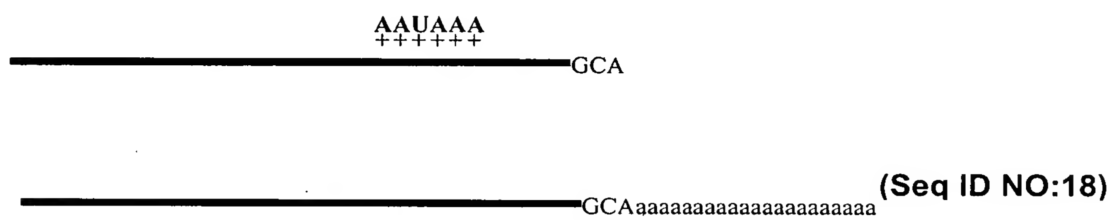
As rescanning documents *will not* correct images, please do not report the images to the Image Problem Mailbox.

Fig. 1 Cleavage and Polyadenylation Process For The SV40 early Poly(A) site

A. CTTATCGATACCGTCGAAACTTGTTTATTGCAGCTTATAATGGTTACAAATAAAGCAATAGCAT
CACAAATTTCACAAATAAAGCATTTTTTTCACTGCATTCTAGTTGTGGTTTGTCCAAACTCATCA
ATGTATCTTATCATGTC **(Seq ID NO:1)** Cleavage site

B.

C.



\+ Upstream and downstream cleavage-polyadenylation elements

(Seq ID NO:2)

Fig 2 E1A transcription control region

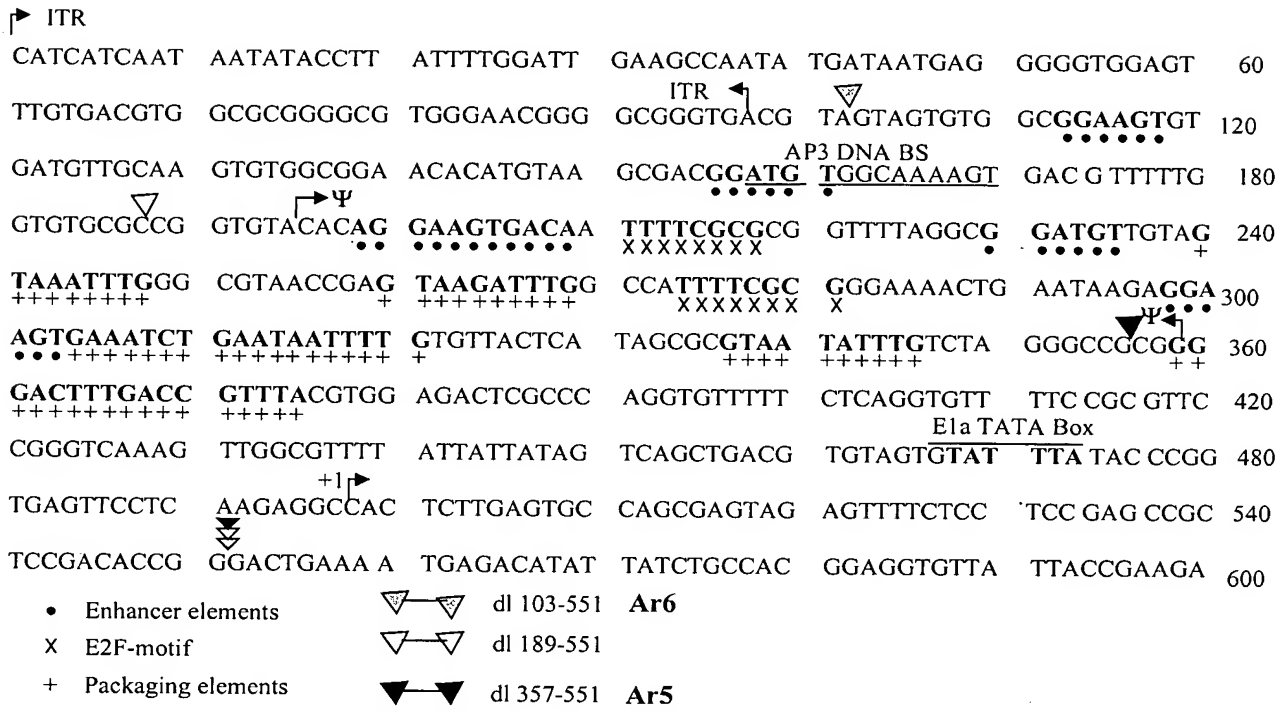


Figure 3. Sequence of Ar6pAE2fF from left and right ends of viral DNA

A. Nucleotides 1-1802 containing ITR, polyA, E2F-1 promoter, E1a and a portion of the E1b gene (Seq ID NO:3)

```
   1  CATCATCAATAATATACCTTATTTTGGATTGAAGCCAATATGATAATGAGGGGGTGGAGT
      +-------------------------ITR------------------------------

  61  TTGTGACGTGGCGCGGGGCGTGGGAACGGGGCGGGTGACGTAGGGCGCGATCAAGCTTAT
      +-------------------------ITR-----------+             +----

 121  CGATACCGTCGAAACTTGTTTATTGCAGCTTATAATGGTTACAAATAAAGCAATAGCATC
      ----------------------------polyA----------------------------

 181  ACAAATTTCACAAATAAAGCATTTTTTTCACTGCATTCTAGTTGTGGTTTGTCCAAACTC
      ----------------------------polyA----------------------------

 241  ATCAATGTATCTTATCATGTCTGGATCCGCGCCGCTAGCGATCATCCGGACAAAGCCTGC
      --------------------+                       +-----------------

 301  GCGCGCCCCGCCCCGCCATTGGCCGTACCGCCCCGCGCCGCCGCCCCATCTCGCCCCTCG
      ------------------------E2F-1 promoter----------------------

 361  CCGCCGGGTCCGGCGCGTTAAAGCCAATAGGAACCGCCGCCGTTGTTCCCGTCACGGCCG
      ------------------------E2F-1 promoter----------------------

 421  GGGCAGCCAATTGTGGCGGCGCTCGGCGGCTCGTGGCTCTTTCGCGGCAAAAAGGATTTG
      ------------------------E2f-1 promoter----------------------

 481  GCGCGTAAAAGTGGCCGGGACTTTGCAGGCAGCGGCGGCCGGGGGCGGAGCGGGATCGAG
      ------------------------E2f-1 promoter----------------------

 541  CCCTCGATGATATCAGATCATCGGATCCCGGTCGACTGAAAATGAGACATATTATCTGCC
      --------------+                     +--------------------------

 601  ACGGAGGTGTTATTACCGAAGAAATGGCCGCCAGTCTTTTGGACCAGCTGATCGAAGAGG
      ------------------------E1a gene--------------------------

 661  TACTGGCTGATAATCTTCCACCTCCTAGCCATTTTGAACCACCTACCCTTCACGAACTGT
      ------------------------E1a gene--------------------------

 721  ATGATTTAGACGTGACGGCCCCCGAAGATCCCAACGAGGAGGCGGTTTCGCAGATTTTTC
      ------------------------E1a gene--------------------------

 781  CCGACTCTGTAATGTTGGCGGTGCAGGAAGGGATTGACTTACTCACTTTTCCGCCGGCGC
      ------------------------E1a gene--------------------------

 841  CCGGTTCTCCGGAGCCGCCTCACCTTTCCCGGCAGCCCGAGCAGCCGGAGCAGAGAGCCT
      ------------------------E1a gene--------------------------

 901  TGGGTCCGGTTTCTATGCCAAACCTTGTACCGGAGGTGATCGATCTTACCTGCCACGAGG
      ------------------------E1a gene--------------------------
```

```
961  CTGGCTTTCCACCCAGTGACGACGAGGATGAAGAGGGTGAGGAGTTTGTGTTAGATTATG
     -------------------------E1a gene-------------------------

1021 TGGAGCACCCCGGGCACGGTTGCAGGTCTTGTCATTATCACCGGAGGAATACGGGGGACC
     -------------------------E1a gene-------------------------

1081 CAGATATTATGTGTTCGCTTTGCTATATGAGGACCTGTGGCATGTTTGTCTACAGTAAGT
     -------------------------E1a gene-------------------------

1141 GAAAATTATGGGCAGTGGGTGATAGAGTGGTGGGTTTGGTGTGGTAATTTTTTTTTTAAT
     -------------------------E1a gene-------------------------

1201 TTTTACAGTTTTGTGGTTTAAAGAATTTTGTATTGTGATTTTTTTAAAAGGTCCTGTGTC
     -------------------------E1a gene-------------------------

1261 TGAACCTGAGCCTGAGCCCGAGCCAGAACCGGAGCCTGCAAGACCTACCCGCCGTCCTAA
     -------------------------E1a gene-------------------------

1321 AATGGCGCCTGCTATCCTGAGACGCCCGACATCACCTGTGTCTAGAGAATGCAATAGTAG
     -------------------------E1a gene-------------------------

1381 TACGGATAGCTGTGACTCCGGTCCTTCTAACACACCTCCTGAGATACACCCGGTGGTCCC
     -------------------------E1a gene-------------------------

1441 GCTGTGCCCCATTAAACCAGTTGCCGTGAGAGTTGGTGGGCGTCGCCAGGCTGTGGAATG
     -------------------------E1a gene-------------------------

1501 TATCGAGGACTTGCTTAACGAGCCTGGGCAACCTTTGGACTTGAGCTGTAAACGCCCCAG
     -------------------------E1a gene-------------------------

1561 GCCATAAGGTGTAAACCTGTGATTGCGTGTGTGGTTAACGCCTTTGTTTGCTGAATGAGT
     -------------------------E1a gene-------------------------

1621 TGATGTAAGTTTAATAAAGGGTGAGATAATGTTTAACTTGCATGGCGTGTTAAATGGGGC
     -------------------------+----------------------------------

1681 GGGGCTTAAAGGGTATATAATGCGCCGTGGGCTAATCTTGGTTACATCTGACCTCATGGA
     -------------------------E1b gene-------------------------

1741 GGCTTGGGAGTGTTTGGAAGATTTTTCTGCTGTGCGTAACTTGCTGGAACAGAGCTCTAA
     -------------------------E1b gene-------------------------

1801 CA
     --
```

B. Nucleotides 33881-34412 containing packaging signal and ITR (Seq ID NO:4)

```
33881 AACCTACGCCCAGAAACGAAAGCCAAAAAACCCACAACTTCCTCAAATCGTCACTTCCGT

33941 TTTCCCACGTTACGTCACTTCCCATTTTAATTAAGAATTCTACAATTCCCAACACATACA
```

```
34001 AGTTACTCCGCCCTAAAACCCTGGGCGAGTCTCCACGTAAACGGTCAAAGTCCCCGCGGC
                          +-packaging signal-----------------------

34061 CCTAGACAAATATTACGCGCTATGAGTAACACAAAATTATTCAGATTTCACTTCCTCTTA
      ---------------------packaging signal-----------------------

34121 TTCAGTTTTCCCGCGAAAATGGCCAAATCTTACTCGGTTACGCCCAAATTTACTACAACA
      ---------------------packaging signal-----------------------

34181 TCCGCCTAAAACCGCGCGAAAATTGTCACTTCCTGTGTACACCGGCGCACACCAAAAACG
      -------------------------------------+

34241 TCACTTTTGCCACATCCGTCGCTTACATGTGTTCCGCCACACTTGCAACATCACACTTCC

34301 GCCACACTACTACGTCACCCGCCCCGTTCCCACGCCCCGCGCCACGTCACAAACTCCACC
                +-----------------ITR------------------------------

34361 CCCTCATTATCATATTGGCTTCAATCCAAAATAAGGTATATTATTGATGATG
      ---------------------------ITR---------------------+
```

Figure 4. Sequence of Ar6F from left end of viral DNA (Seq ID NO:5)

```
1 CATCATCAATAATATACCTTATTTTGGATTGAAGCCAATATGATAATGAGGGGGTGGAGT
      +-------------------------ITR---------------------------------

 61 TTGTGACGTGGCGCGGGGCGTGGGAACGGGGCGGGTGACGTAGGGCGCGCCGCTAGCGAT
     ---------------------------ITR----------------++------MCS--------

121 ATCGGATCCCGGTCGACTGAAAATGAGACATATTATCTGCCACGGAGGTGTTATTACCGA
    --------------+------------E1a-------------------------------

181 AGAAATGGCCGCCAGTCTTTTGGACCAGCTGATCGAAGAGGTACTGGCTGATAATCTTCC
    ----------------------------E1a-------------------------------

241 ACCTCCTAGCCATTTTGAACCACCTACCCTTCACGAACTGTATGATTTAGACGTGACGGC
    ----------------------------E1a-------------------------------

301 CCCCGAAGATCCCAACGAGGAGGCGGTTTCGCAGATTTTTCCCGACTCTGTAATGTTGGC
    ----------------------------E1a-------------------------------

361 GGTGCAGGAAGGGATTGACTTACTCACTTTTCCGCCGGCGCCCGGTTCTCCGGAGCCGCC
    ----------------------------E1a-------------------------------

421 TCACCTTTCCCGGCAGCCCGAGCAGCCGGAGCAGAGAGCCTTGGGTCCGGTTTCTATGCC
    ----------------------------E1a-------------------------------

481 AAACCTTGTACCGGAGGTGATCGATCTTACCTGCCACGAGGCTGGCTTTCCACCCAGTGA
    ----------------------------E1a-------------------------------

541 CGACGAGGATGAAGAGGGTGAGGAGTTTGTGTTAGATTATGTGGAGCACCCCGGGCACGG
    ----------------------------E1a-------------------------------

601 TTGCAGGTCTTGTCATTATCACCGGAGGAATACGGGGGACCCAGATATTATGTGTTCGCT
    ----------------------------E1a-------------------------------
```

Figure 5. Sequence of Ar6pAF from left end of viral DNA (Seq ID NO:6)

```
   1 CATCATCAATAATATACCTTATTTTGGATTGAAGCCAATATGATAATGAGGGGGTGGAGT
     +-------------------------ITR---------------------------------

  61 TTGTGACGTGGCGCGGGGCGTGGGAACGGGGCGGGTGACGTAGGGCGCGATCAAGCTTAT
     ---------------------------ITR--------------+            +----

 121 CGATACCGTCGAAACTTGTTTATTGCAGCTTATAATGGTTACAAATAAAGCAATAGCATC
     ---------------------------polyA-------------------------------

 181 ACAAATTTCACAAATAAAGCATTTTTTTCACTGCATTCTAGTTGTGGTTTGTCCAAACTC
     ---------------------------polyA-------------------------------

 241 ATCAATGTATCTTATCATGTCTGGATCCGCGCCGCTAGCGATATCGGATCCCGGTCGACT
     ----------------------+                                      +--

 301 GAAAATGAGACATATTATCTGCCACGGAGGTGTTATTACCGAAGAAATGGCCGCCAGTCT
     ---------------------------E1a--------------------------------

 361 TTTGGACCAGCTGATCGAAGAGGTACTGGCTGATAATCTTCCACCTCCTAGCCATTTTGA
     ---------------------------E1a--------------------------------

 421 ACCACCTACCCTTCACGAACTGTATGATTTAGACGTGACGGCCCCCGAAGATCCCAACGA
     ---------------------------E1a--------------------------------

 481 GGAGGCGGTTTCGCAGATTTTTCCCGACTCTGTAATGTTGGCGGTGCAGGAAGGGATTGA
     ---------------------------E1a--------------------------------

 541 CTTACTCACTTTTCCGCCGGCGCCCGGTTCTCCGGAGCCGCCTCACCTTTCCCGGCAGCC
     ---------------------------E1a--------------------------------

 601 CGAGCAGCCGGAGCAGAGAGCCTTGGGTCCGGTTTCTATGCCAAACCTTGTACCGGAGGT
     ---------------------------E1a--------------------------------
```

Figure 6. Schematic diagram of Ar6pAF and Ar6pAE2fF vectors

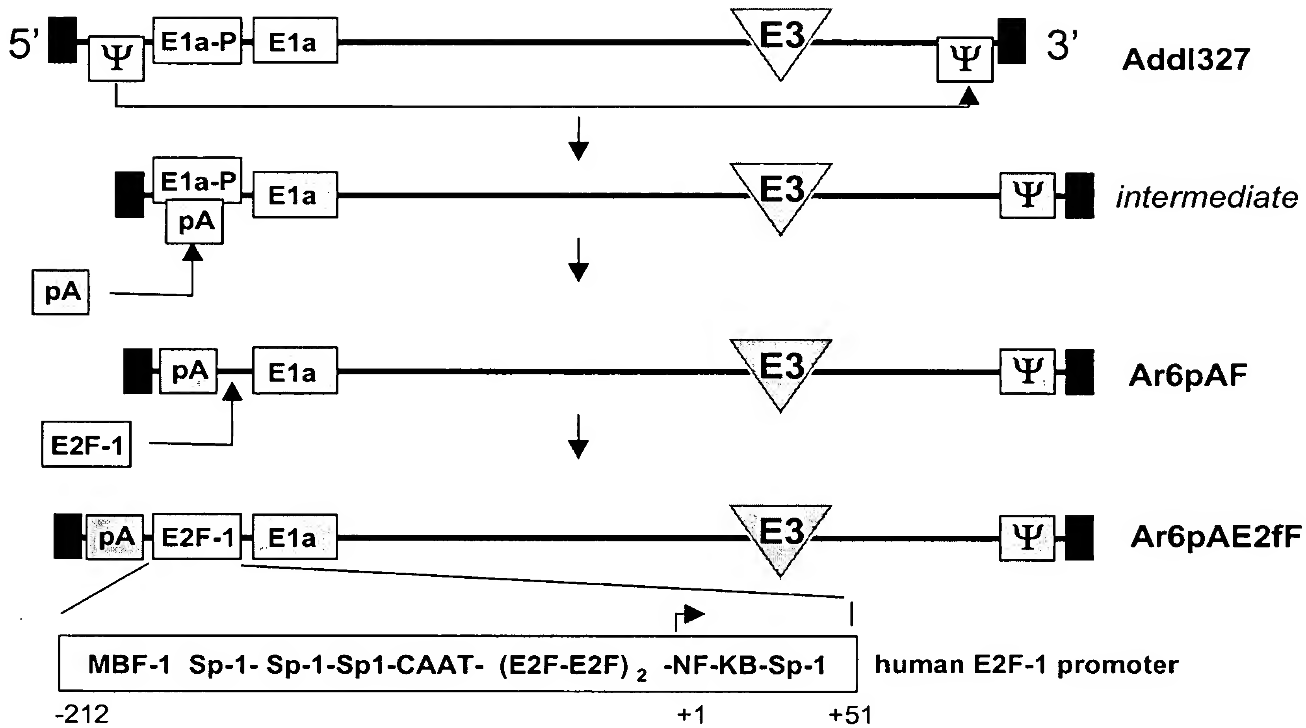


Fig. 7 Body weight change

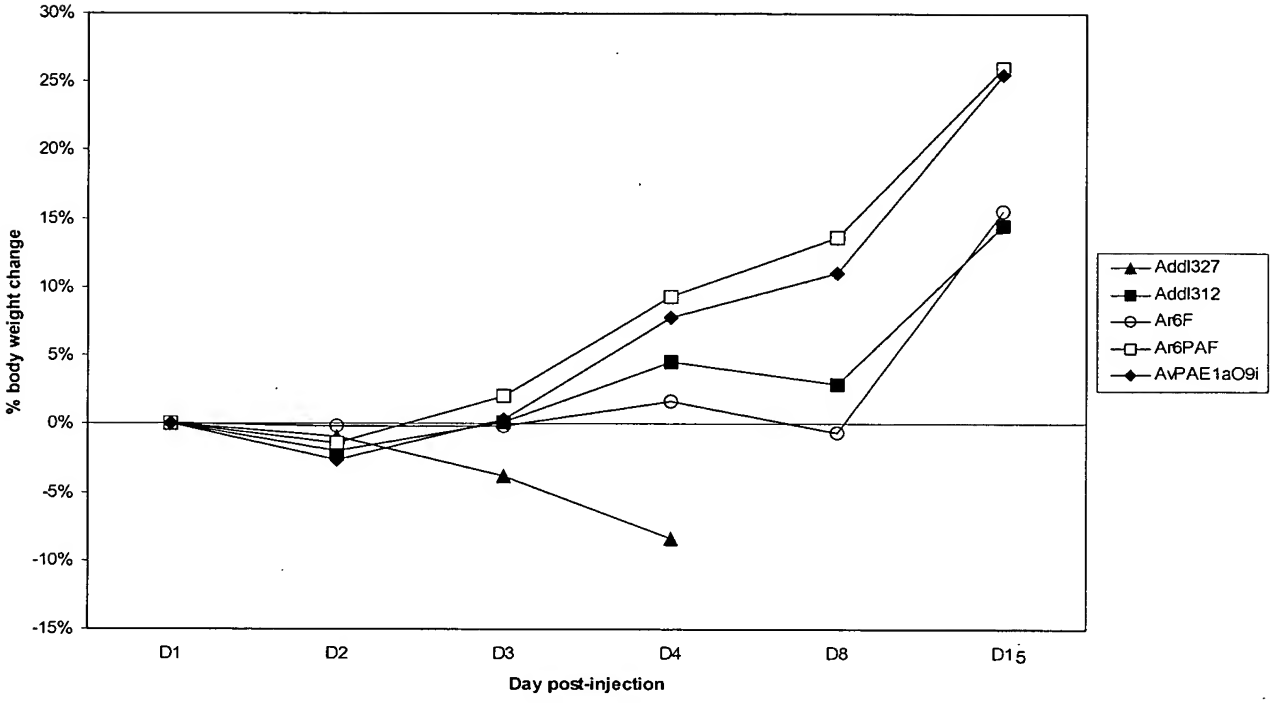


Fig. 8 Minimizing nonspecific transactivation of E1a gene

Backbones generated:

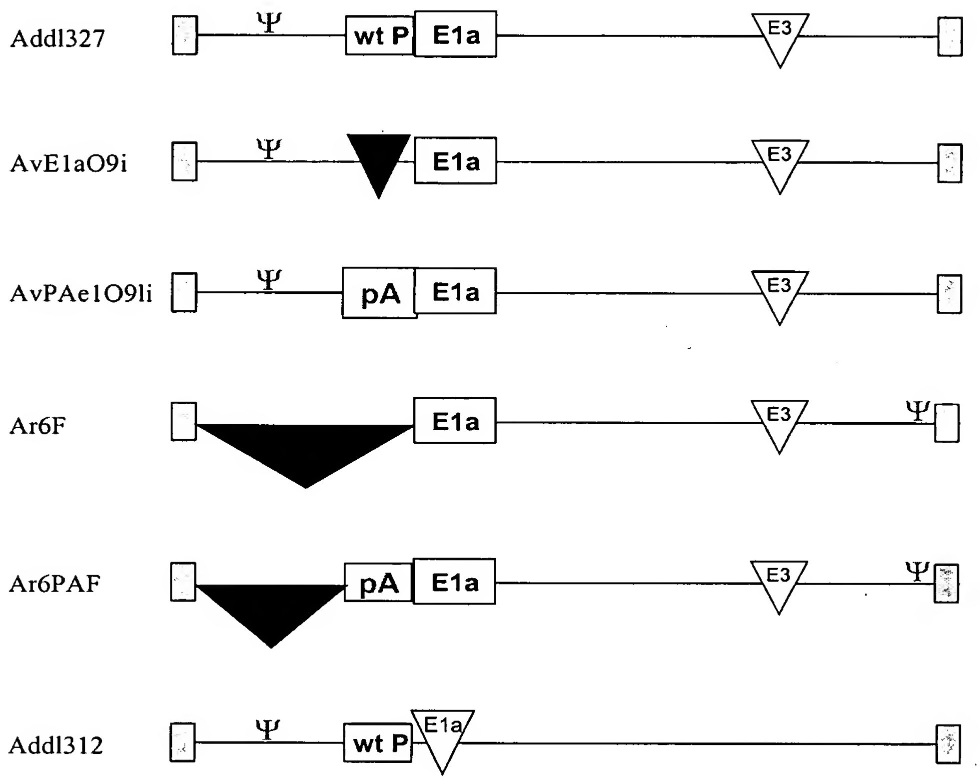


Figure 9. Mean H460 tumor volume

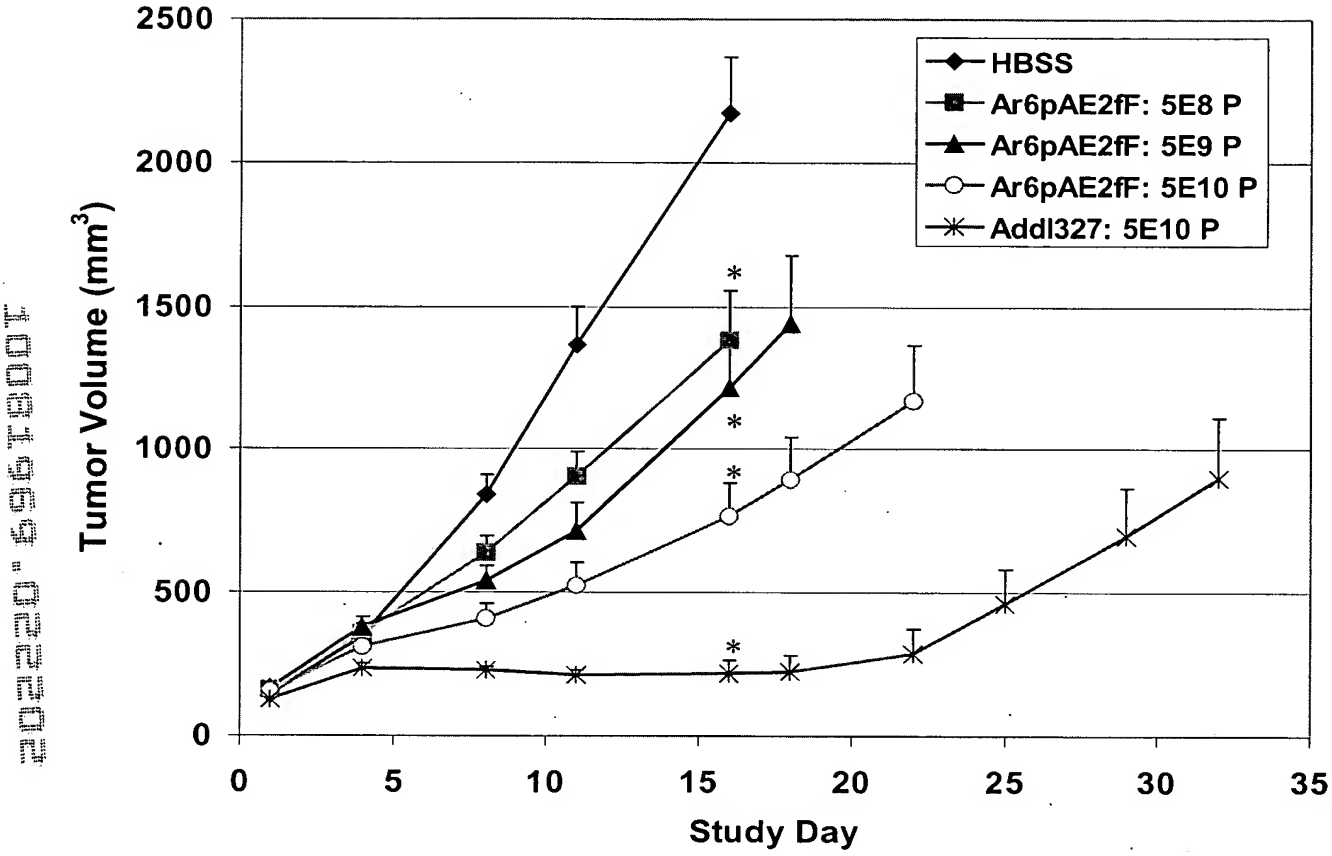
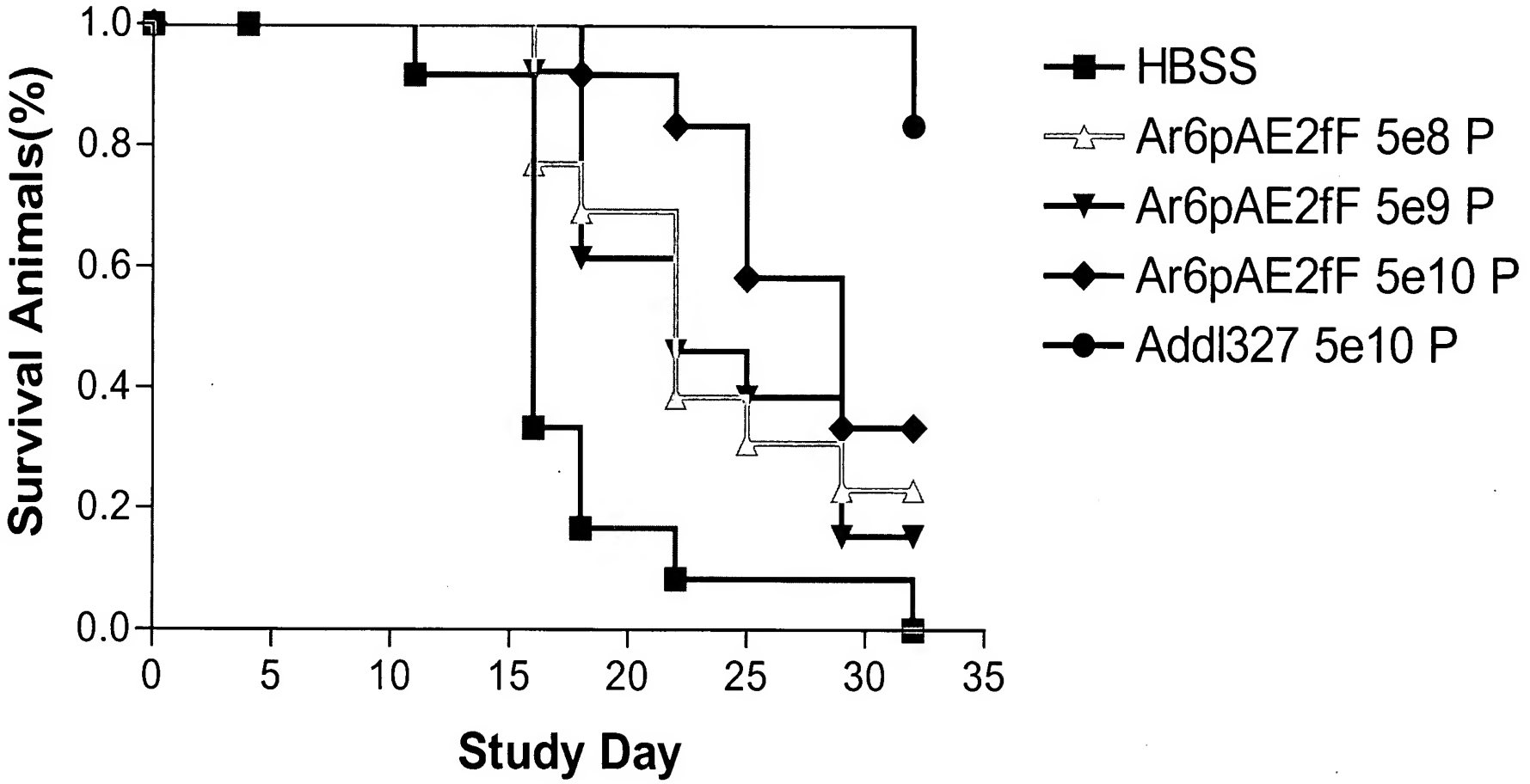
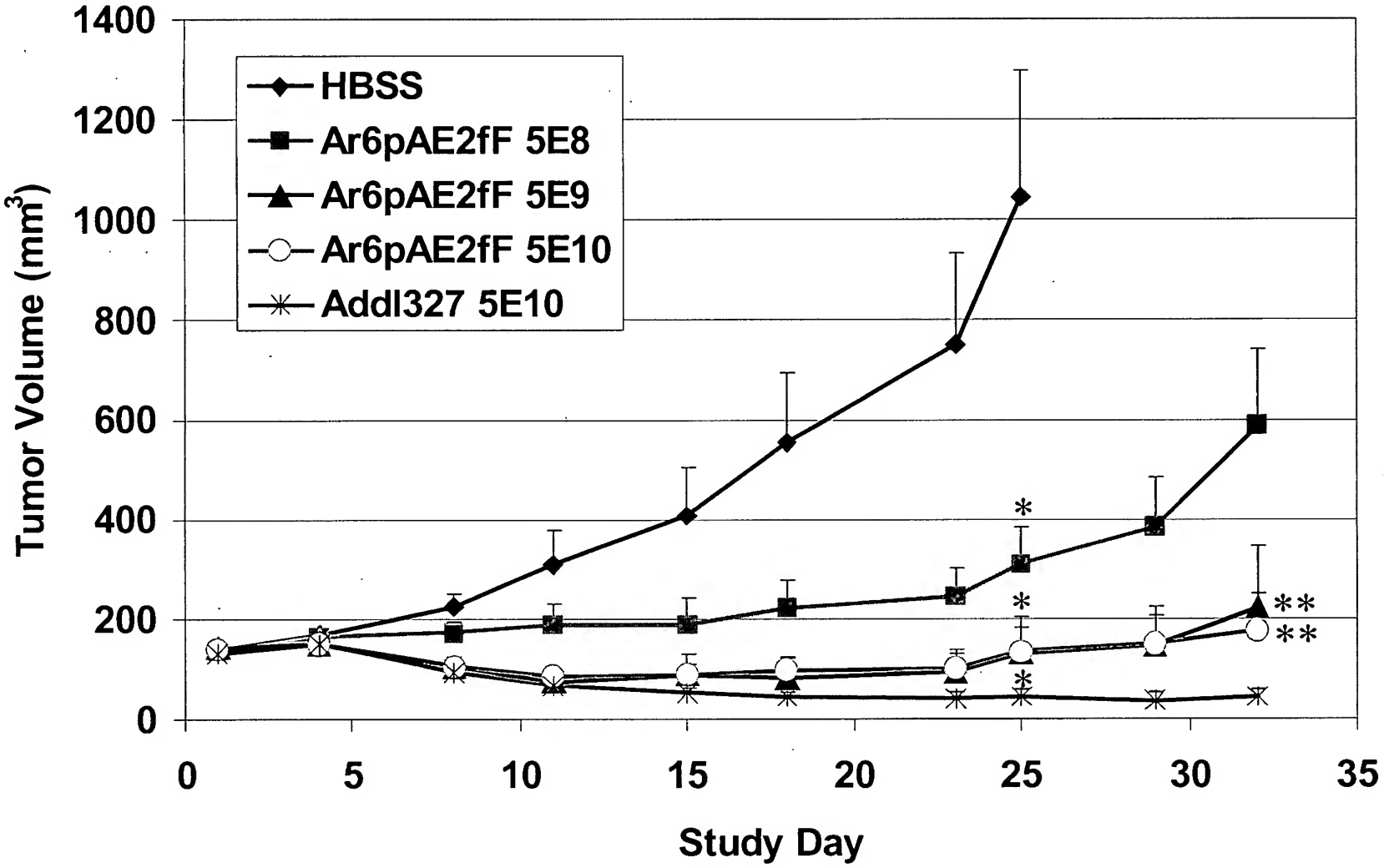


Figure 10. Survival following intratumoral administration of vectors to H460 tumors



igure 11. Mean Hep3B tumor volumes



igure 12. Survival following intratumoral administration of vector to Hep3B tumors

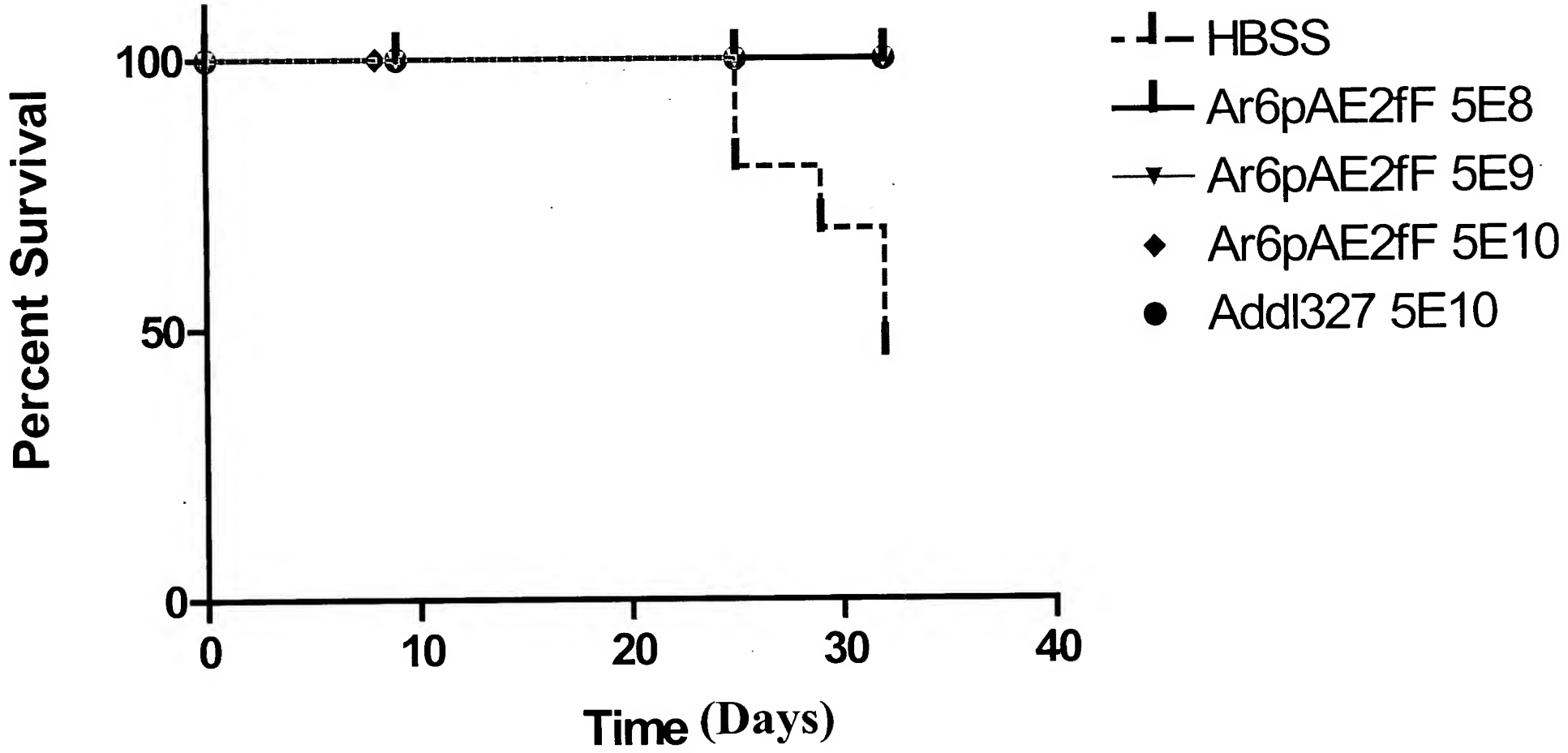


Figure 13. Schematic diagram of adenovirus right donor plasmid pDR2F.

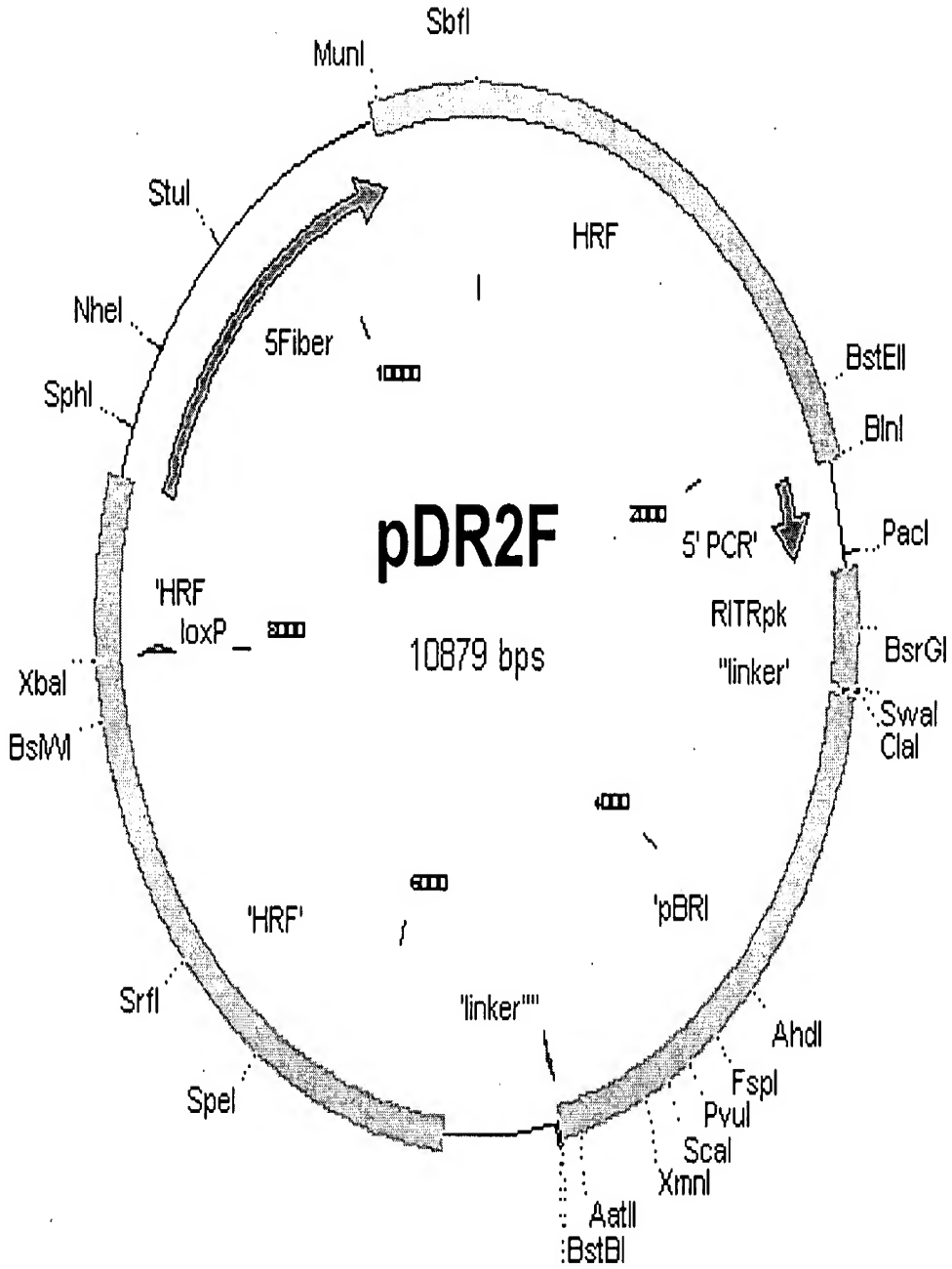


Figure 14. Schematic diagram of adenovirus right donor plasmid pDR2mGmF.

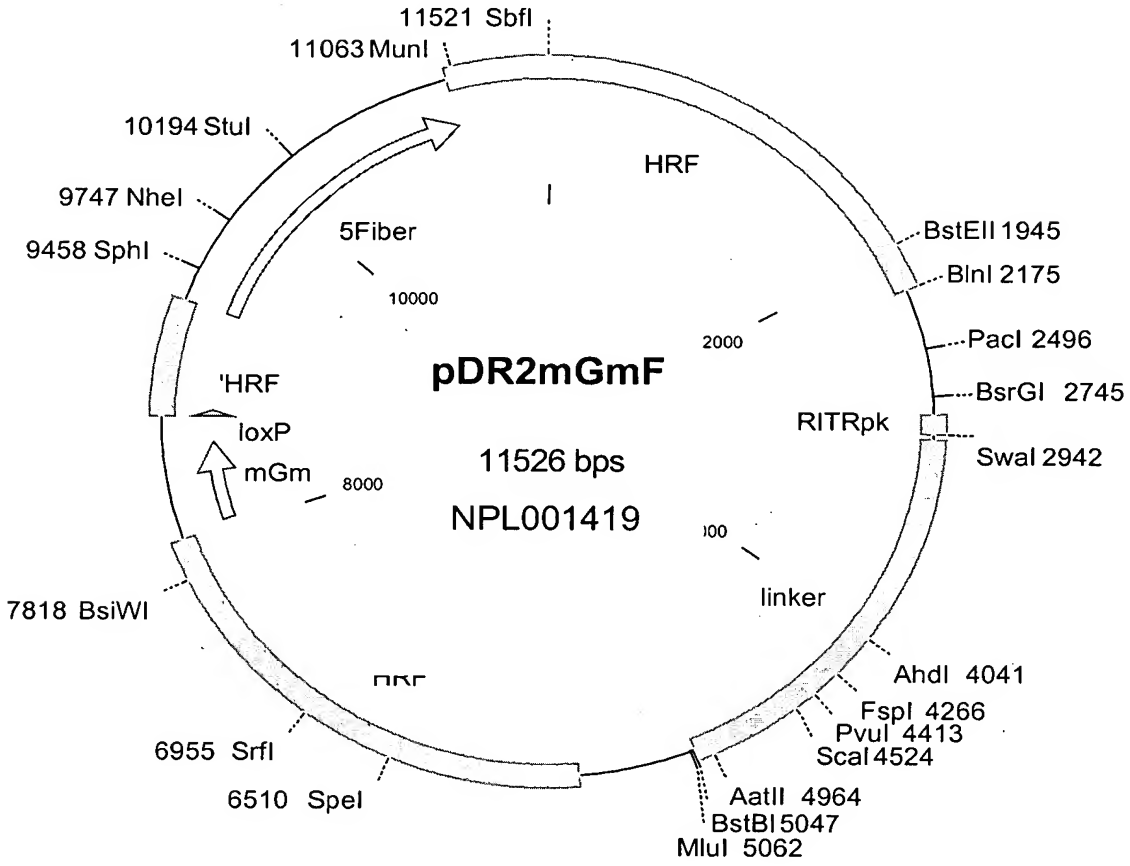


Figure 15. Schematic diagram of plasmid pG1mGmSvNa.

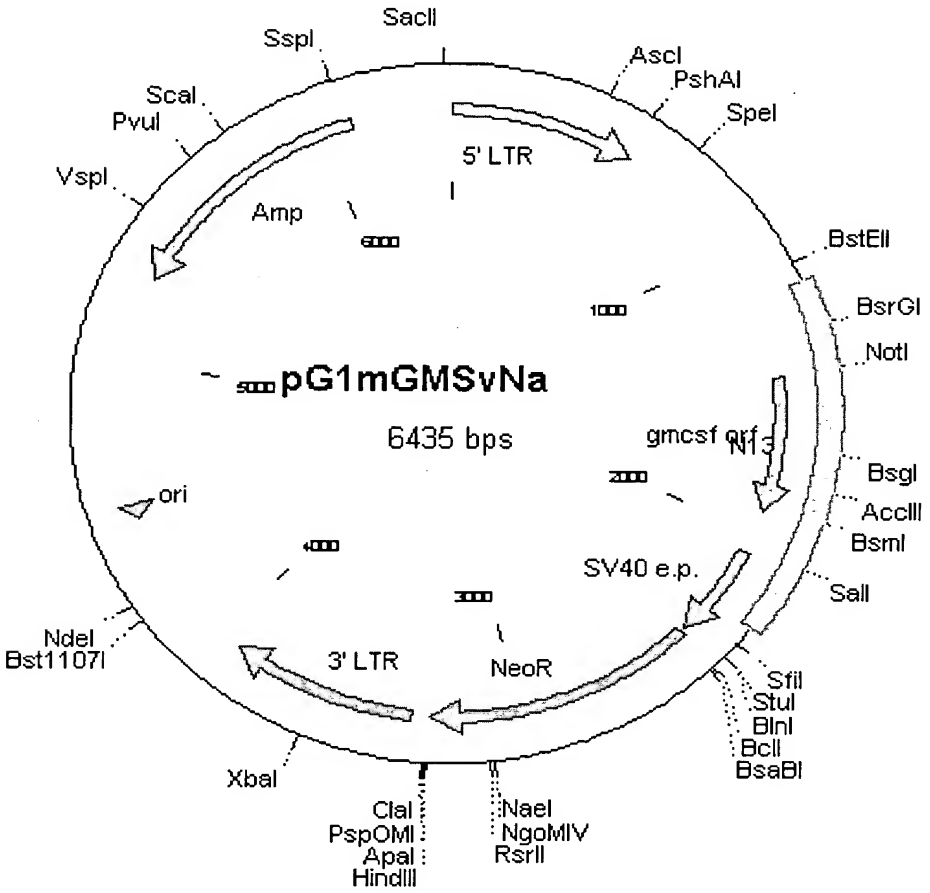


Figure 16. Schematic diagram of plasmid pG1NaSvBg.

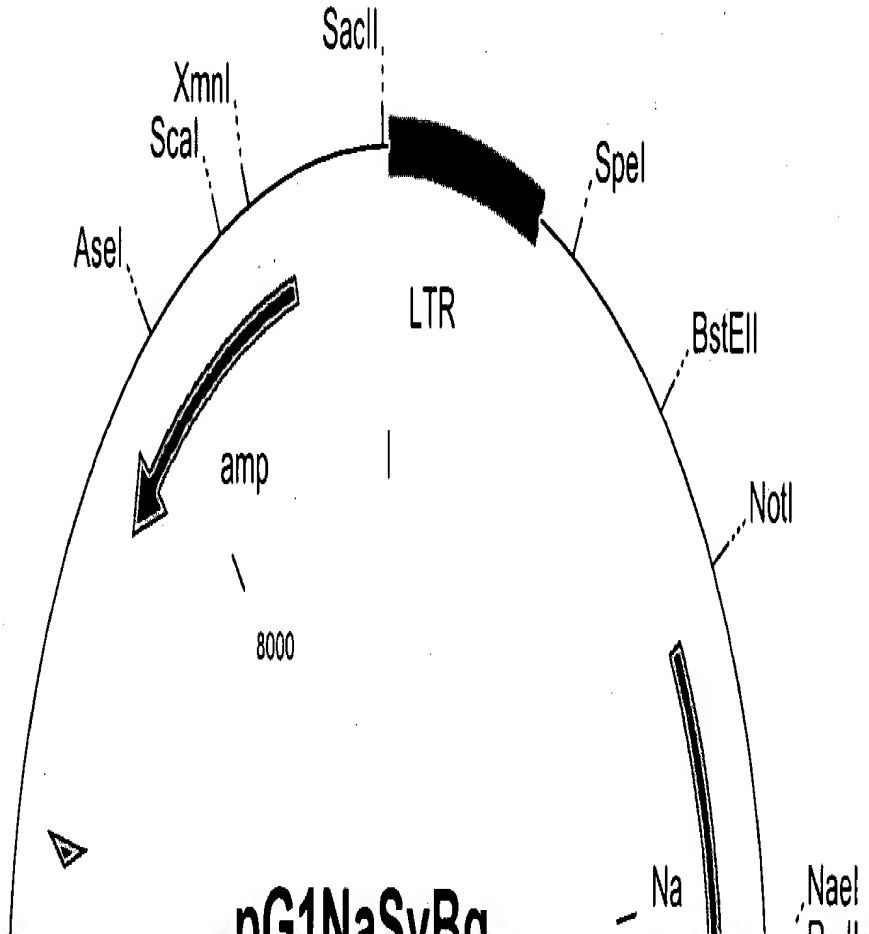


Figure 17. Sequence of the murine GM-CSF cDNA (Seq ID NO:7) and protein (Seq ID NO:8).

```
7878 TTCCGGACAG ACCTCAATAA CTCTGTTTAC CAGAACAGGA GGTGAGCTTA
7928 GAAAACCCTT AGGGTATTAG GCCAAAGGCG CAGCTACTGT GGGGTTTATG
7978 AACAATTCAA GCAACTCTAC GGGCTATTCT AATTCAGGTT TCTCTAGCCG
8028 GGCTGCAGGA ATTCGATGGC CGCTACCTAC AATGGCCCAC GAGAGAAAGG
                                        M  A  H   E  R  K
8078 CTAAGGTCCT GAGGAGGATG TGGCTGCAGA ATTTACTTTT CCTGGGCATT
       A  K  V   L  R  R  M   W  L  Q   N  L  L   F  L  G  I
8128 GTGGTCTACA GCCTCTCAGC ACCCACCCGC TCACCCATCA CTGTCACCCG
        V  V  Y   S  L  S   A  P  T  R   S  P  I   T  V  T
8178 GCCTTGGAAG CATGTAGAGG CCATCAAAGA AGCCCTGAAC CTCCTGGATG
      R  P  W  K   H  V  E   A  I  K   E  A  L  N   L  L  D
8228 ACATGCCTGT CACATTGAAT GAAGAGGTAG AAGTCGTCTC TAACGAGTTC
       D  M  P   V  T  L  N   E  E  V   E  V  V   S  N  E  F
8278 TCCTTCAAGA AGCTAACATG TGTGCAGACC CGCCTGAAGA TATTCGAGCA
        S  F  K   K  L  T   C  V  Q  T   R  L  K   I  F  E
8328 GGGTCTACGG GGCAATTTCA CCAAACTCAA GGGCGCCTTG AACATGACAG
      Q  G  L  R   G  N  F   T  K  L   K  G  A  L   N  M  T
8378 CCAGCTACTA CCAGACATAC TGCCCCCCAA CTCCGGAAAC GGACTGTGAA
       A  S  Y   Y  Q  T  Y   C  P  P   T  P  E   T  D  C  E
8428 ACACAAGTTA CCACCTATGC GGATTTCATA GACAGCCTTA AAACCTTTCT
        T  Q  V   T  T  Y   A  D  F  I   D  S  L   K  T  F
8478 GACTGATATC CCCTTTGAAT GCAAAAAACC AGTCCAAAAA TGAGGAAGCC
      L  T  D  I   P  F  E   C  K  K   P  V  Q  K   -
8528 CAGGCCAGCT CTGAATCCAG CTTCTCAGAC TGCTGCTTTT GTGCCTGCGT
8578 AATGAGCCAG GAACTCGGAA TTTCTGCCTT AAAGGGACCA AGAGATGTGG
8628 CACAGGTAGT CGAATCAAGC TTATCGATAC CGTCGACCTC GACTAGATAA
8678 CTTCGTATAA TGTATGCTAT ACGAAGTTAT GCTAGAAATG GACGGAATTA
8728 TTACAGAGCA GCGCCTGCTA GAAAGACGCA GGGCAGCGGC CGAGCAACAG
8778 CGCATGAATC AAGAGCTCCA AGACATGGTT AACTTGCACC AGTGCAAAA 8826
```

Figure 18. Pathway used to generate pAr6pAE2fmGmF plasmid.

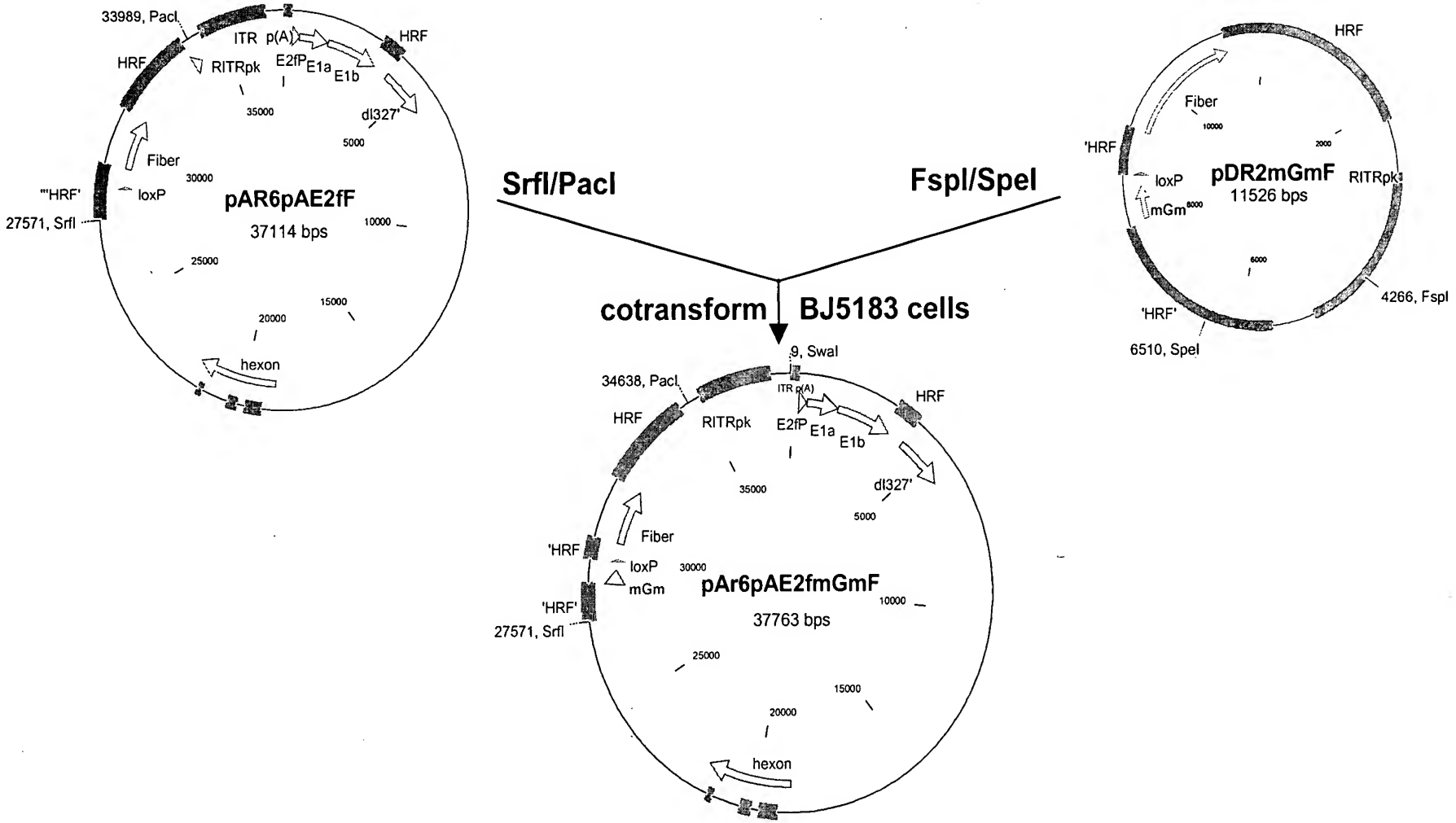


Figure 19. MTS assay of oncolytic vectors on different tumor cell lines.

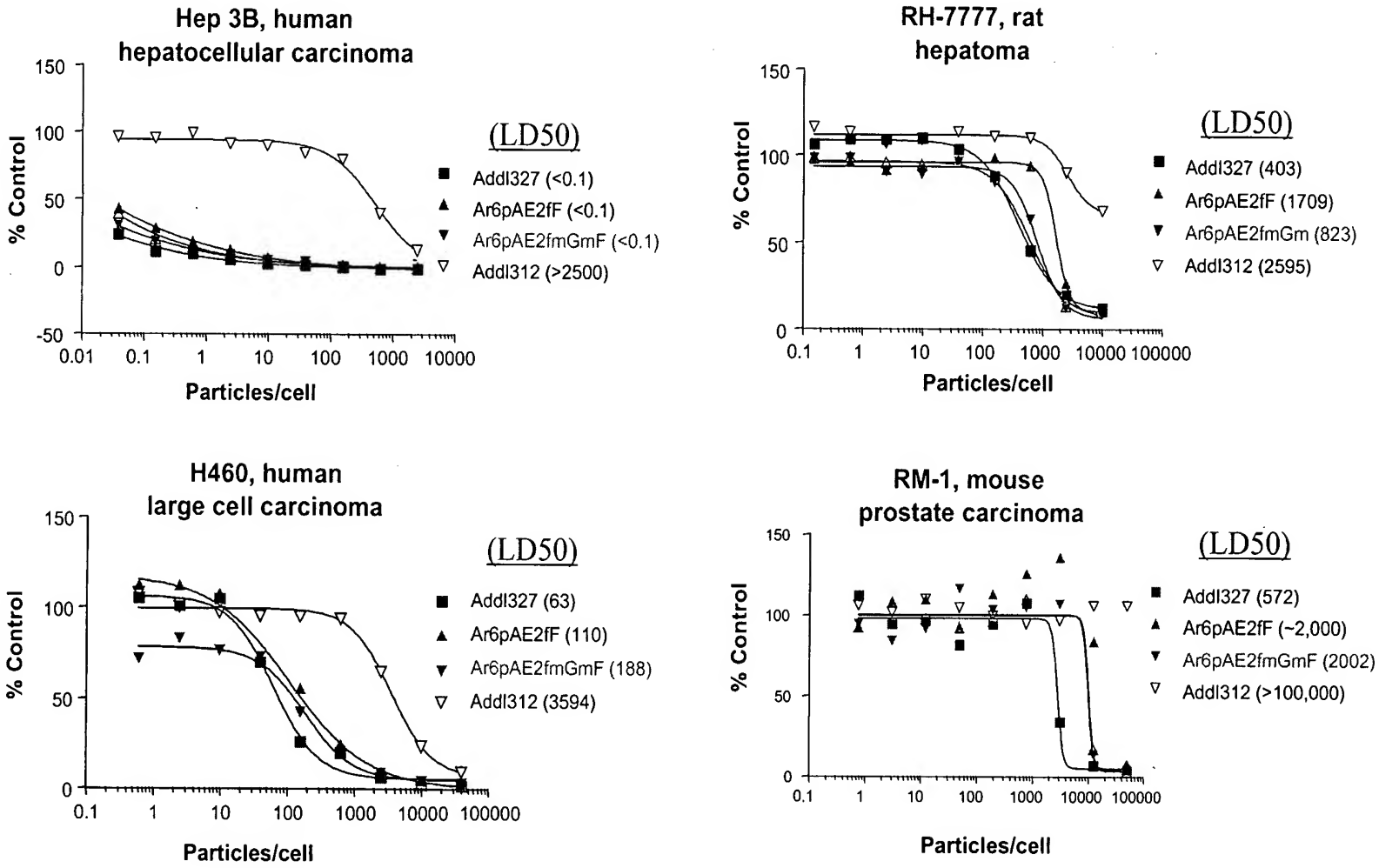


Figure 20. Sequence of the human GM-CSF cDNA (Seq ID NO:19) and protein (Seq ID NO:20).

```
28536  TATTAGGCCA AAGGCGCAGC TACTGTGGGG TTTATGAACA ATTCAAGCAA
28586  CTCTACGGGC TATTCTAATT CAGGTTTCTC TAGGATCTTT CCGCAGCAGC

28636  CGCCACCATG TGGCTGCAGA GCCTGCTGCT CTTGGGCACT GTGGCCTGCA
                  M  W  L  Q  S  L  L  L  L  G  T  V  A  C

28686  GCATCTCTGC ACCCGCCCGC TCGCCCAGCC CCAGCACGCA GCCCTGGGAG
        S  I  S  A  P  A  R  S  P  S  P  S  T  Q  P  W  E

28736  CATGTGAATG CCATCCAGGA GGCCCGGCGT CTCCTGAACC TGAGTAGAGA
        H  V  N  A  I  Q  E  A  R  R  L  L  N  L  S  R

28786  CACTGCTGCT GAGATGAATG AAACAGTAGA AGTCATCTCA GAAATGTTTG
       D  T  A  A  E  M  N  E  T  V  E  V  I  S  E  M  F

28836  ACCTCCAGGA GCCGACCTGC CTACAGACCC GCCTGGAGCT GTACAAGCAG
        D  L  Q  E  P  T  C  L  Q  T  R  L  E  L  Y  K  Q

28886  GGCCTGCGGG GCAGCCTCAC CAAGCTCAAG GGCCCCTTGA CCATGATGGC
         G  L  R  G  S  L  T  K  L  K  G  P  L  T  M  M

28936  CAGCCACTAC AAGCAGCACT GCCCTCCAAC CCCGGAAACT TCCTGTGCAA
       A  S  H  Y  K  Q  H  C  P  P  T  P  E  T  S  C  A

28986  CCCAGACTAT CACCTTTGAA AGTTTCAAAG AGAACCTGAA GGACTTTCTG
        T  Q  T  I  T  F  E  S  F  K  E  N  L  K  D  F  L

29036  CTTGTCATCC CCTTTGACTG CTGGGAGCCA GTCCAGGAGT GAGTCGACAA
         L  V  I  P  F  D  C  W  E  P  V  Q  E  -

29086  GCTCTAGATA ACTTCGTATA ATGTATGCTA TACGAAGTTA TGCTAGAAAT
29136  GGACGGAATT ATTACAGAGC AGCGCCTGCT AGAAAGACGC AGGGCAGCGG
29186  CCGAGCAACA GCGCATGAAT CAAGAGCTCC AAGACATGGT TAACTTGCAC
29236  CAGTGCAAAA GGGGTATCTT TTGTCTGGTA AAGCAGG  29273
```

gure 21. Pathway used to generate pAr6pAE2fhGmF plasmid.

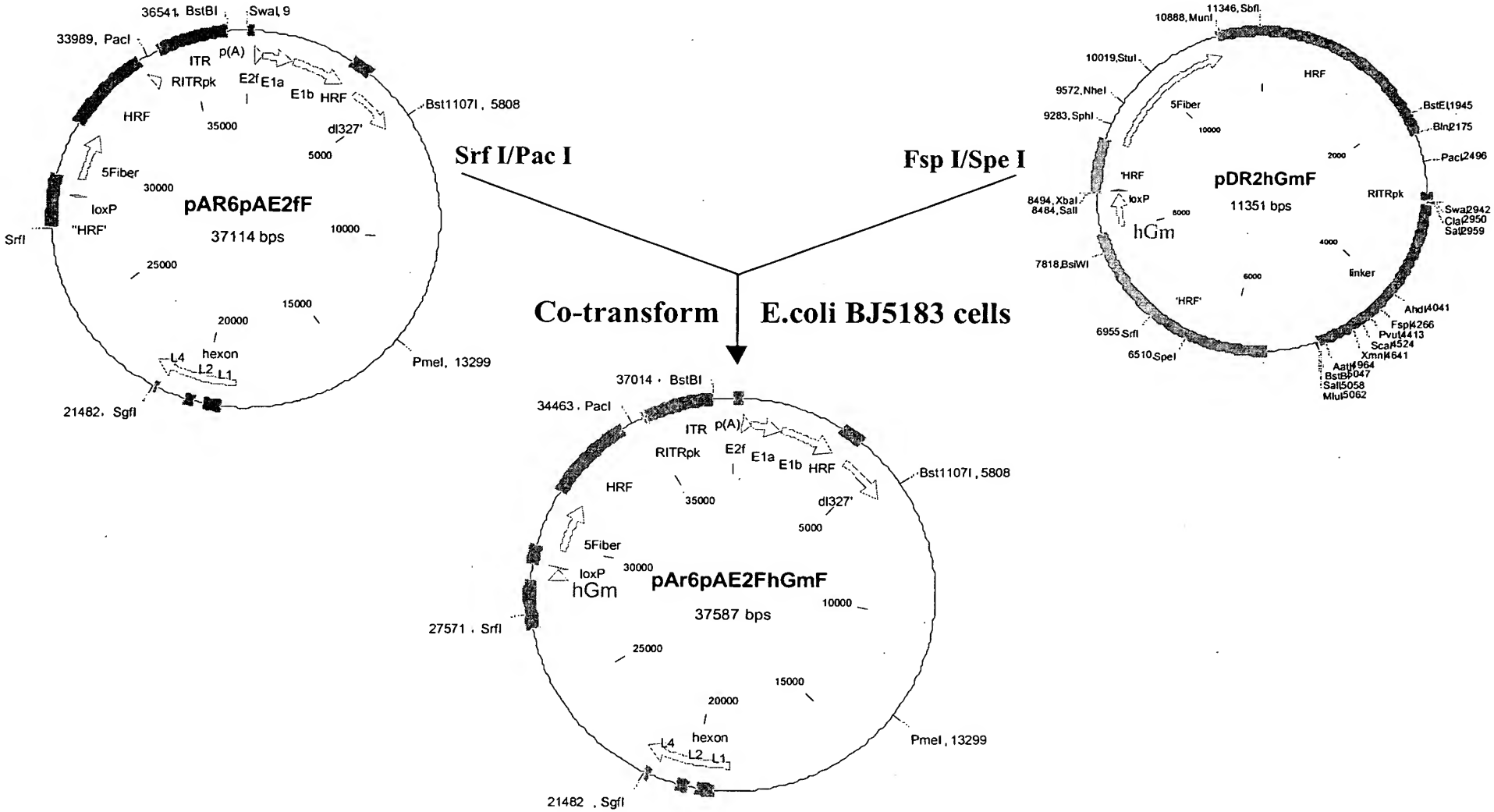


Figure 22. MTS assay of oncolytic vectors on different tumor cell lines.

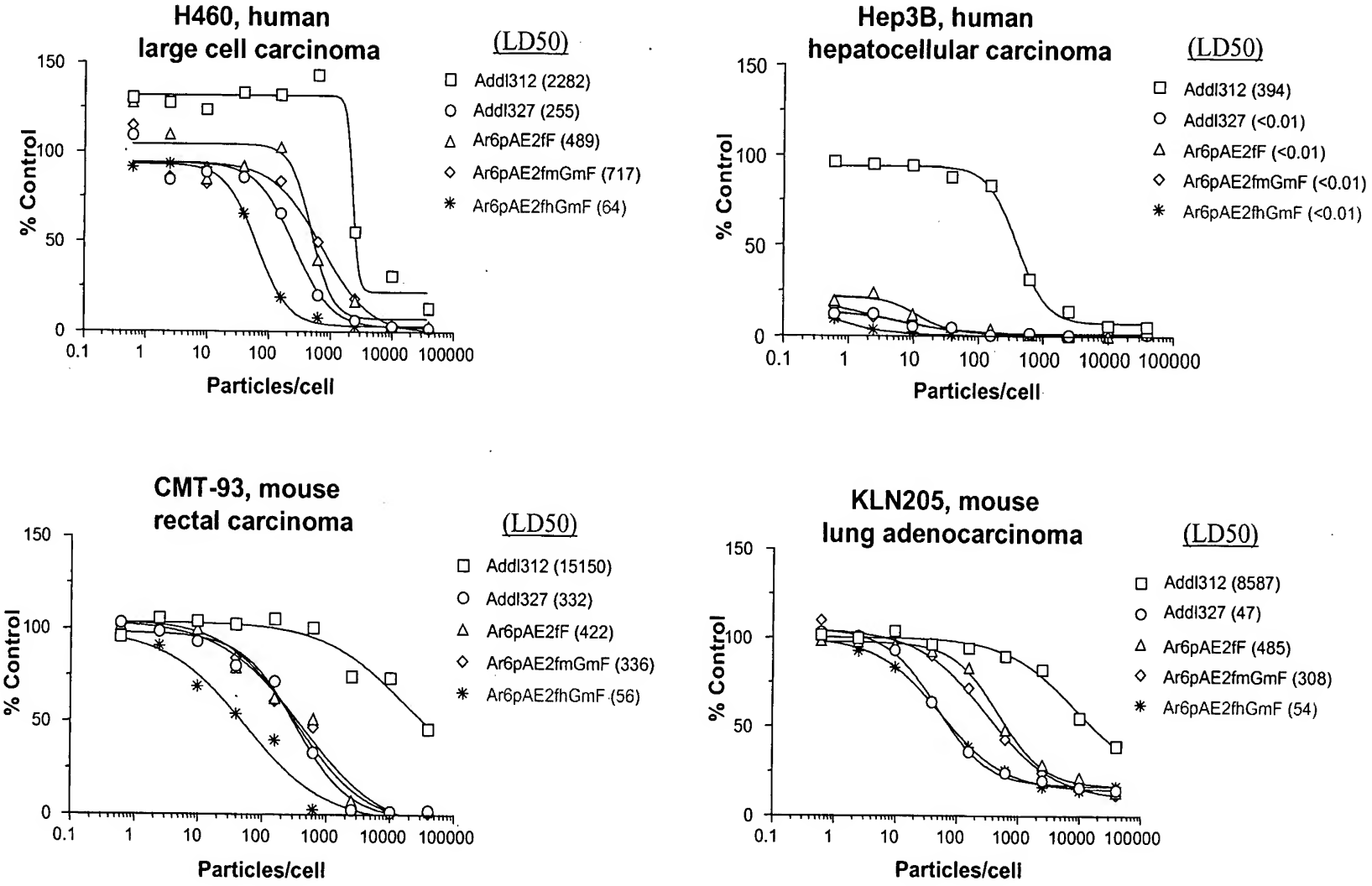


Figure 23. Efficacy of GM-CSF armed oncolytic vectors in H460 tumor model

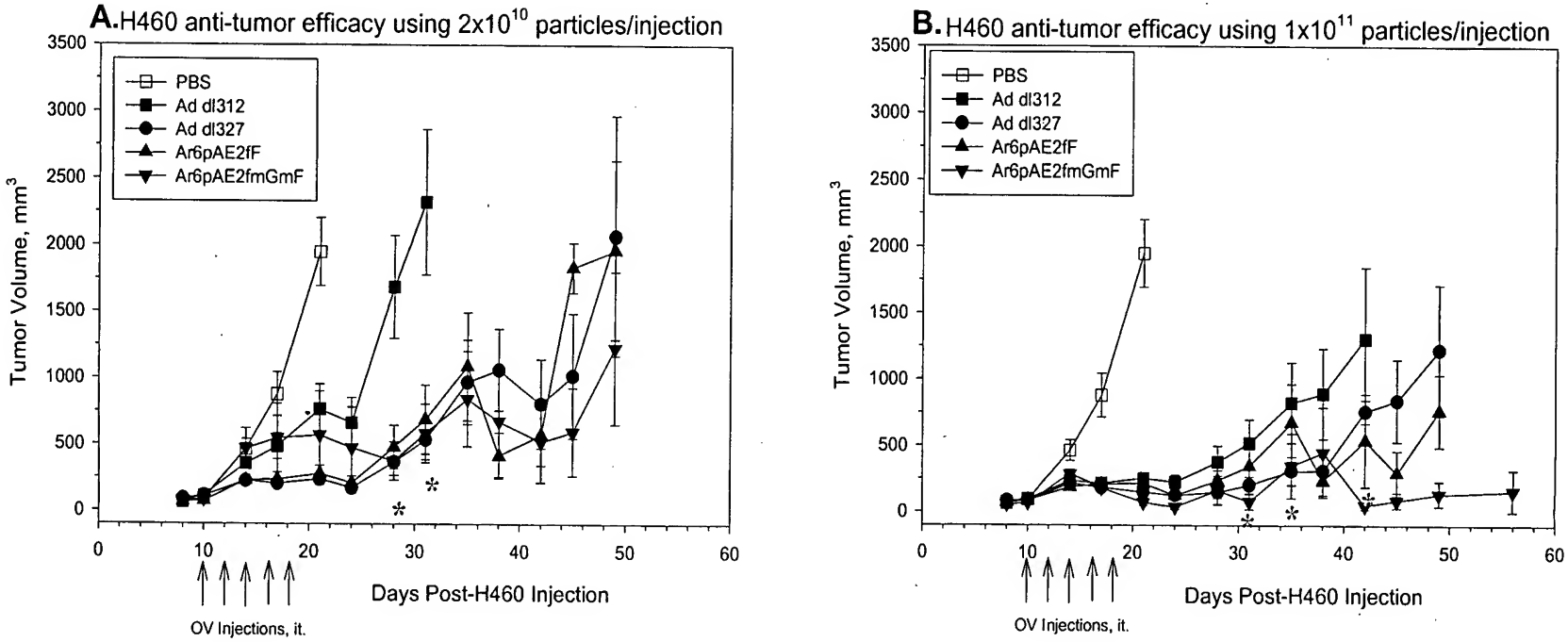


Figure 24. Efficacy of GM-CSF armed oncolytic vectors in Hep3B tumor model

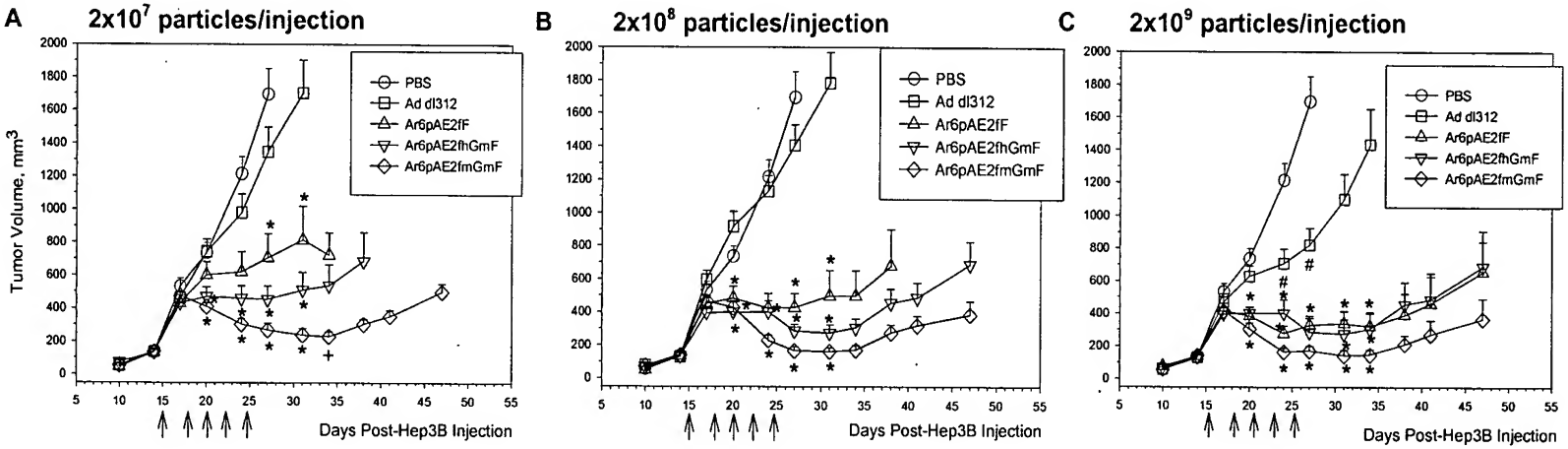


Figure 25. Schematic Diagram of PCR and Overlap PCR for Δgp19 Donor Plasmids

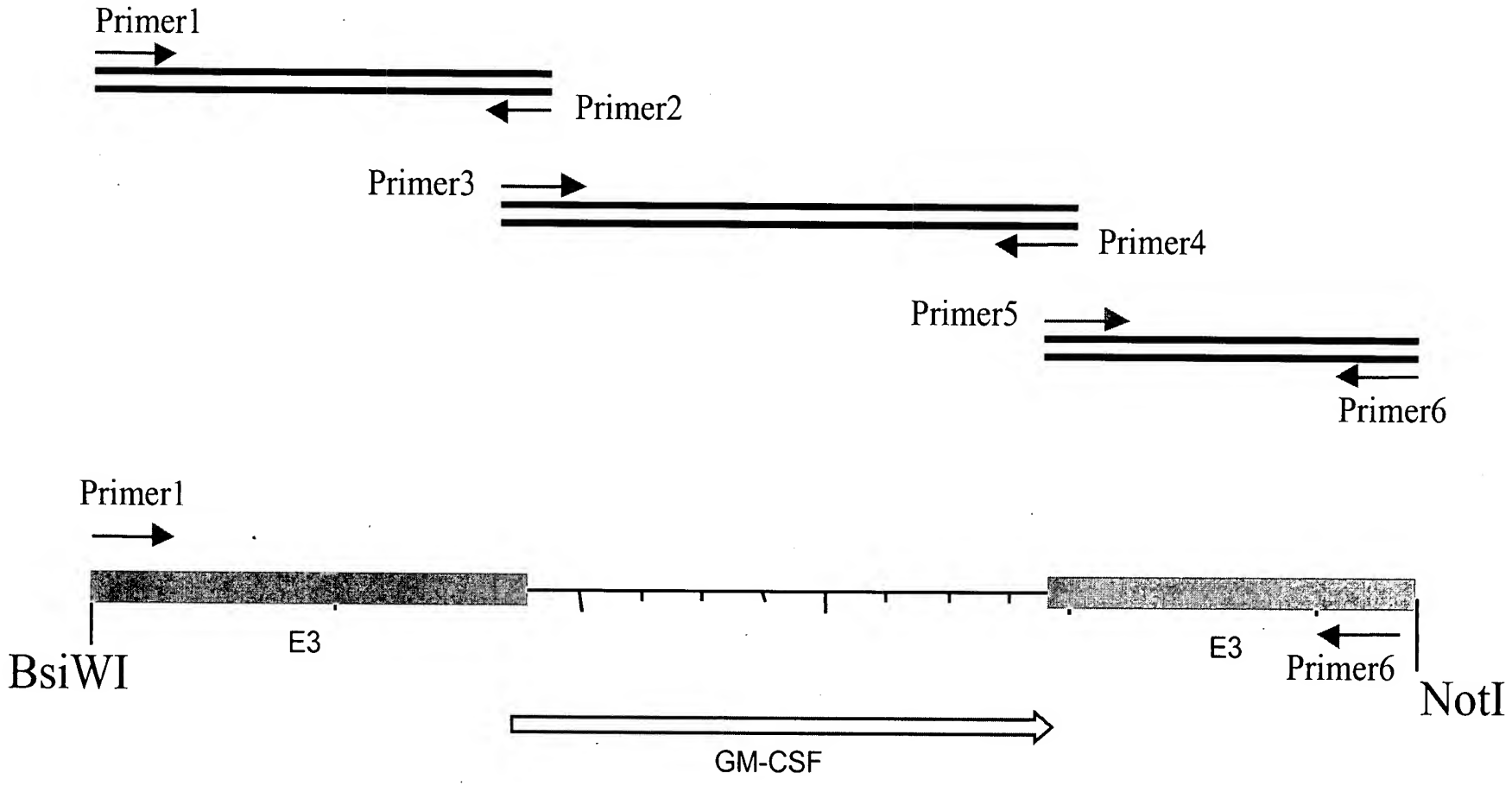
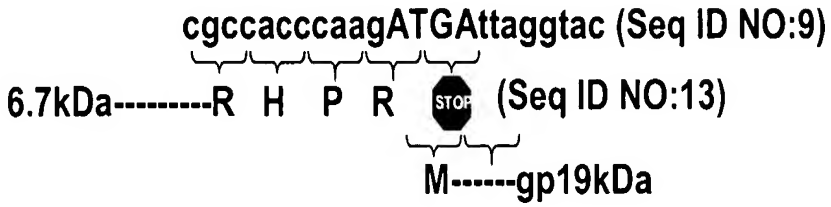


Figure 26. Schematic Diagram of □gp19 Vectors

a. Sequence of native E3 region:



b. Sequence comparison of Δgp19 vectors at the junction between E3-6.7 and GM-CSF:

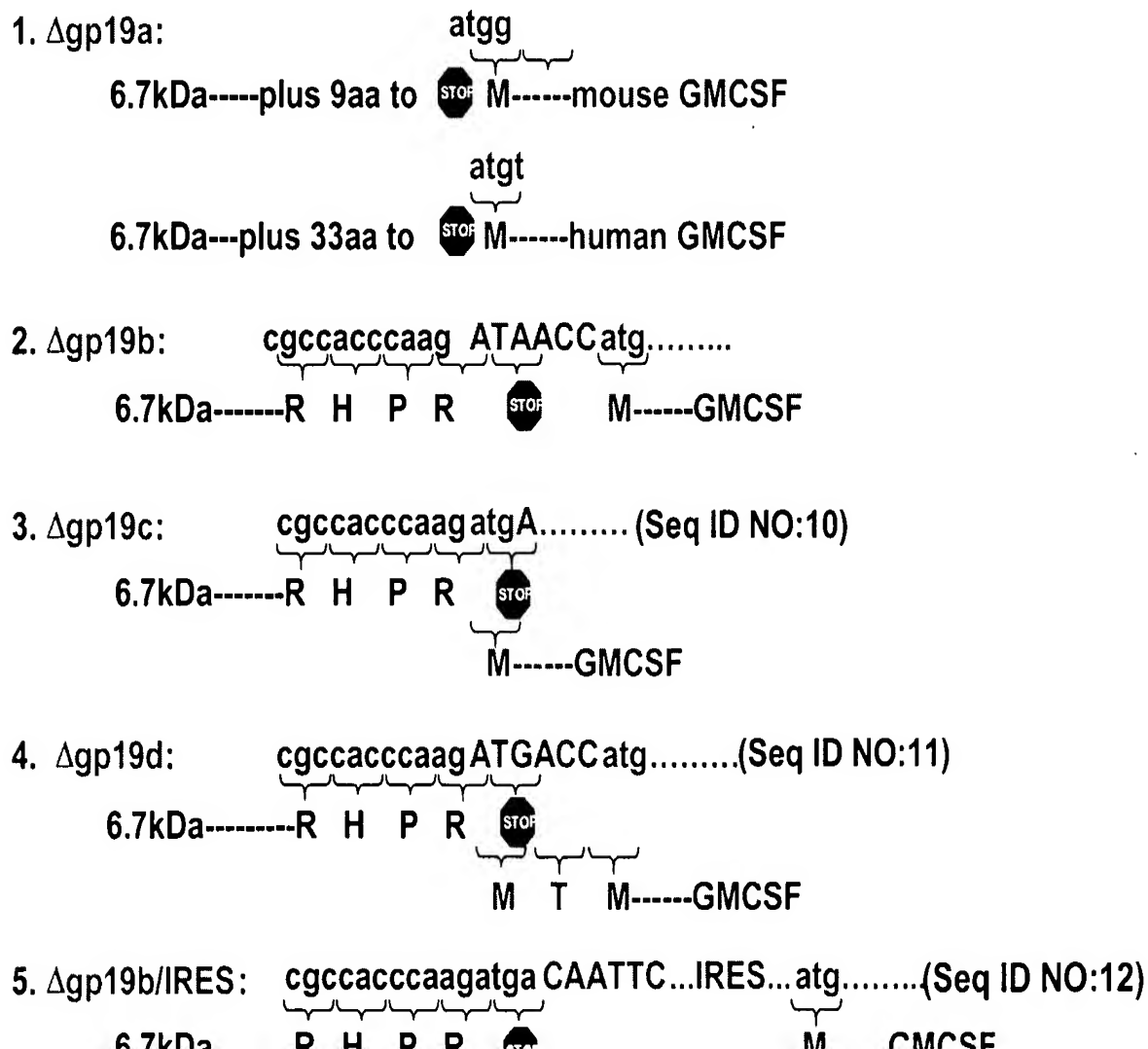


Figure 27a. Pathway Used to Generate the pAr6pAE2f(E3+,mGm,Dg19b)F Large Plasmid

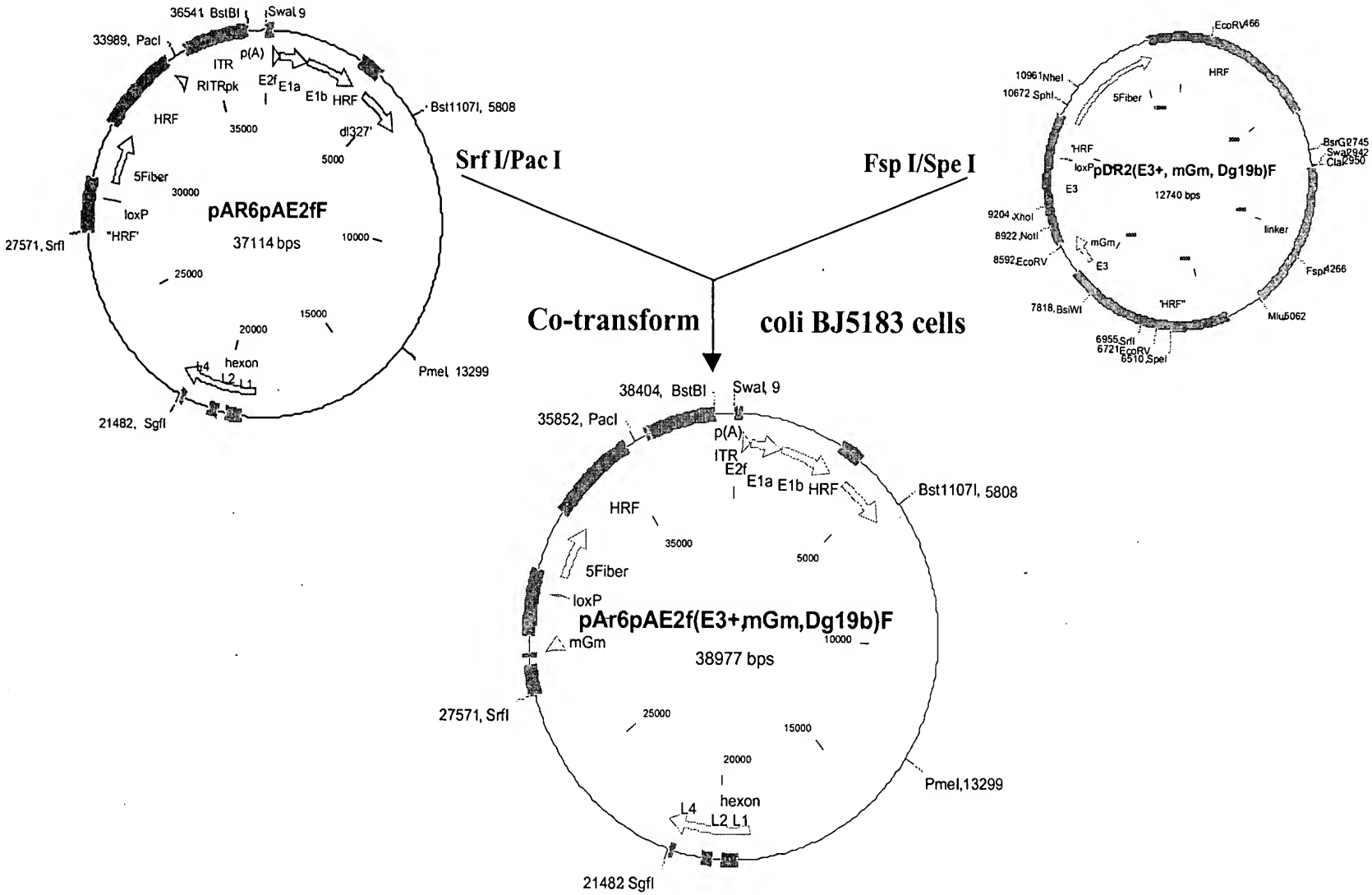


Figure 27b. Pathway Used to Generate the pAr6pAE2f(E3+,hGm,Dg19b)F Large Plasmid

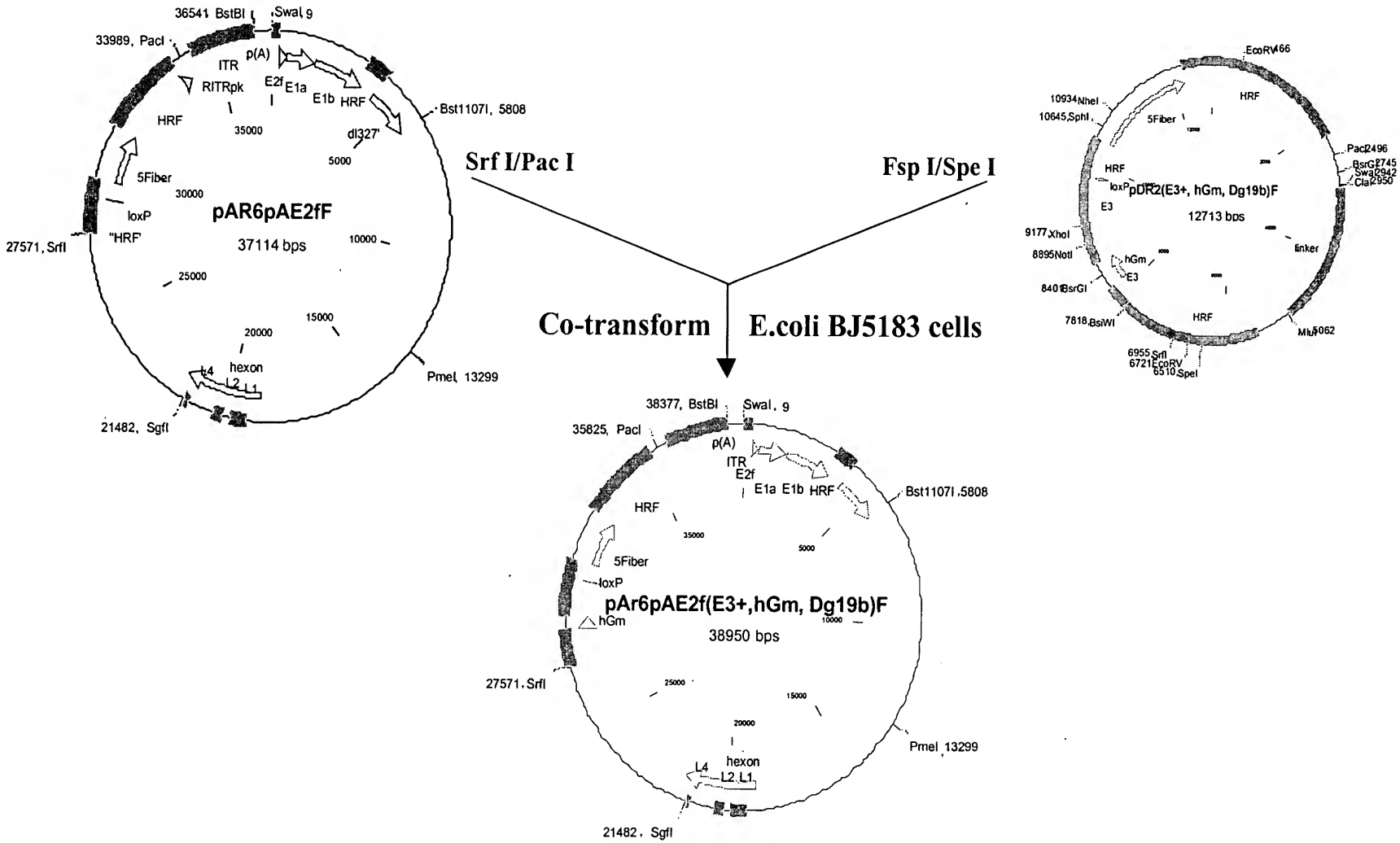
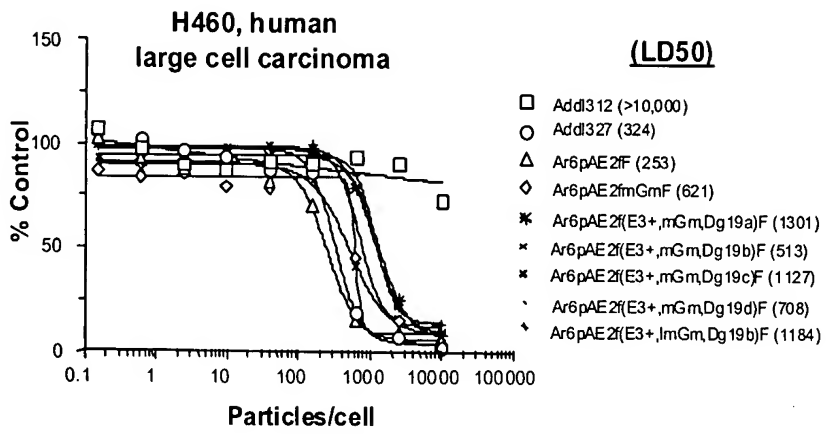
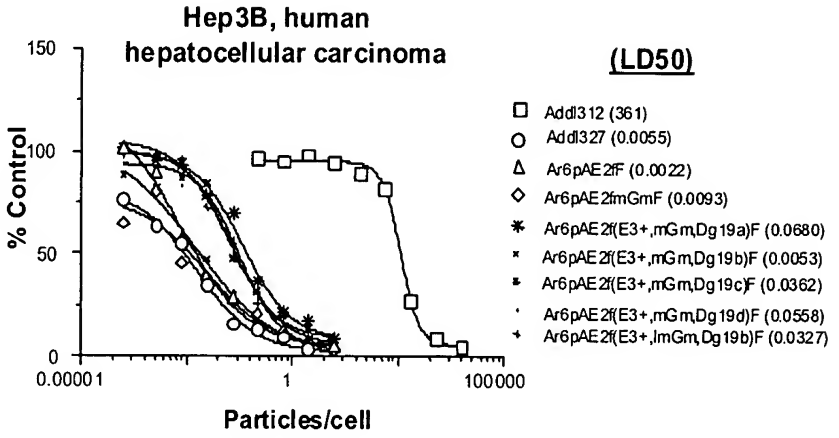


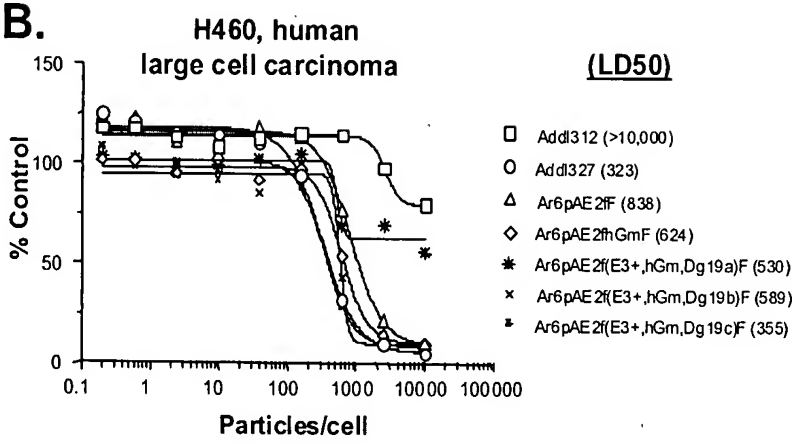
Figure 28. MTS Assay of Δgp19 mGM-CSF Vectors on H460 and Hep3B Tumor Cell Lines

A.





B.



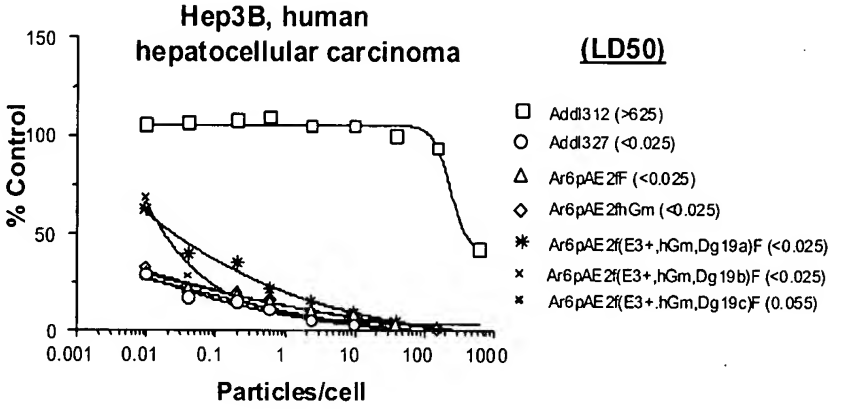
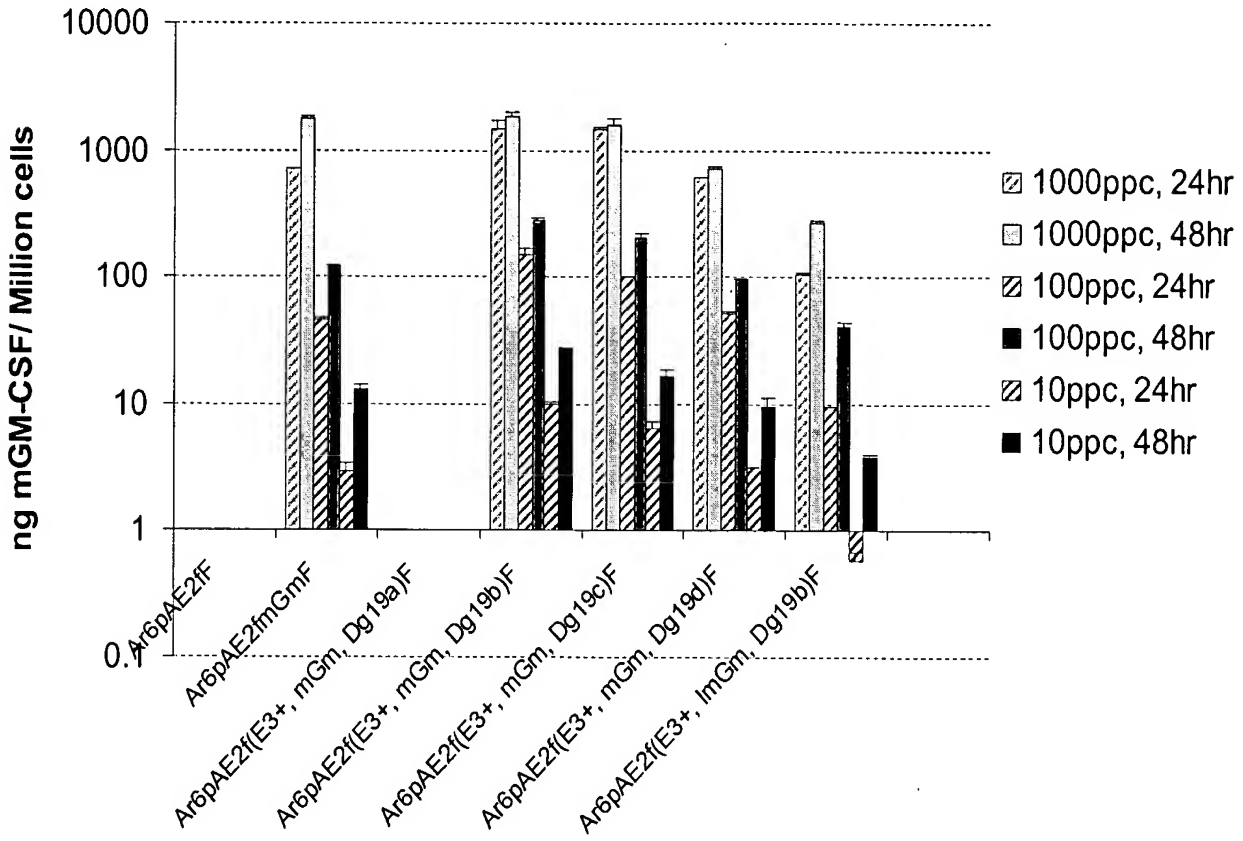


Figure 29. GM-CSF Expression Mediated by Δgp19 GM-CSF Vectors in Infected H460 Cells Detected by ELISA

a. Mouse GM-CSF expression in H460 cells



b. Human GM-CSF expression in H460 cells

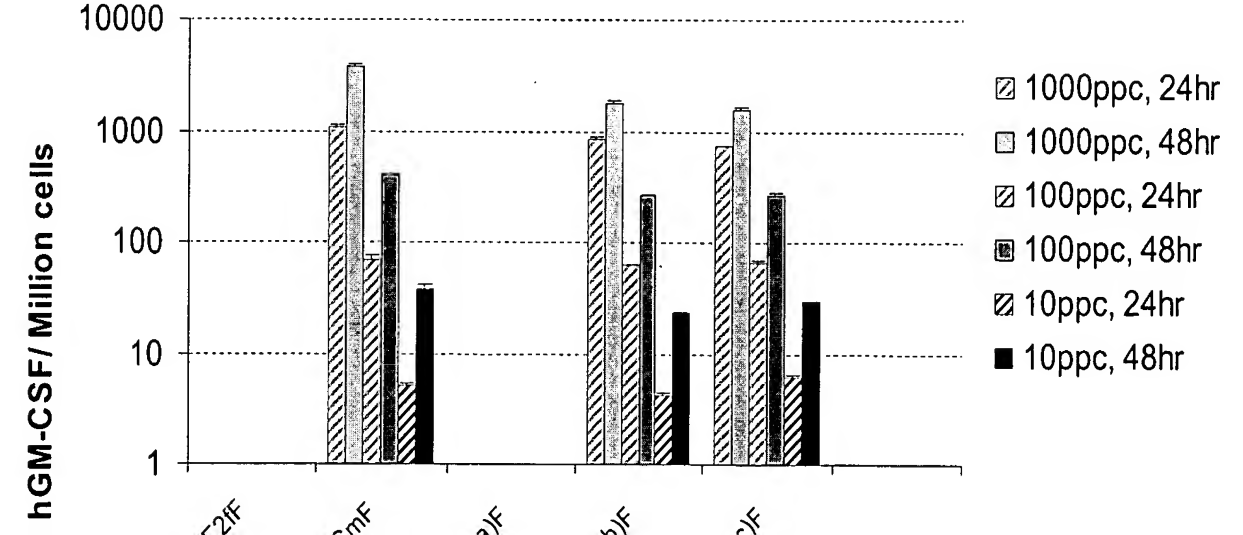


Figure 30. Anti-Tumor Activity of Oncolytic Adenoviruses ($2x10^9$ particles/injection) in the Hep3B Xenograft Subcutaneous Tumor Model

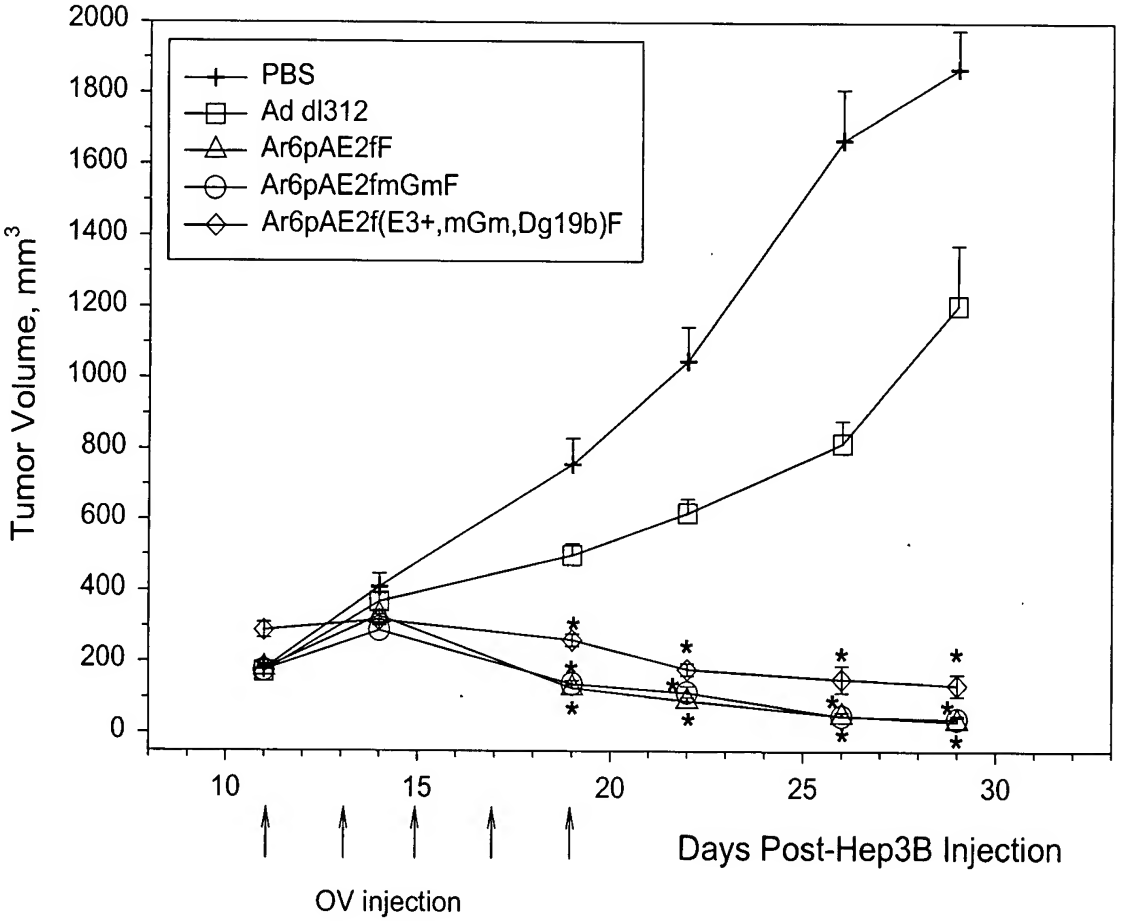


Figure 31. Anti-Tumor Activity of Oncolytic Adenoviruses in the H460 Xenograft Subcutaneous Tumor Model

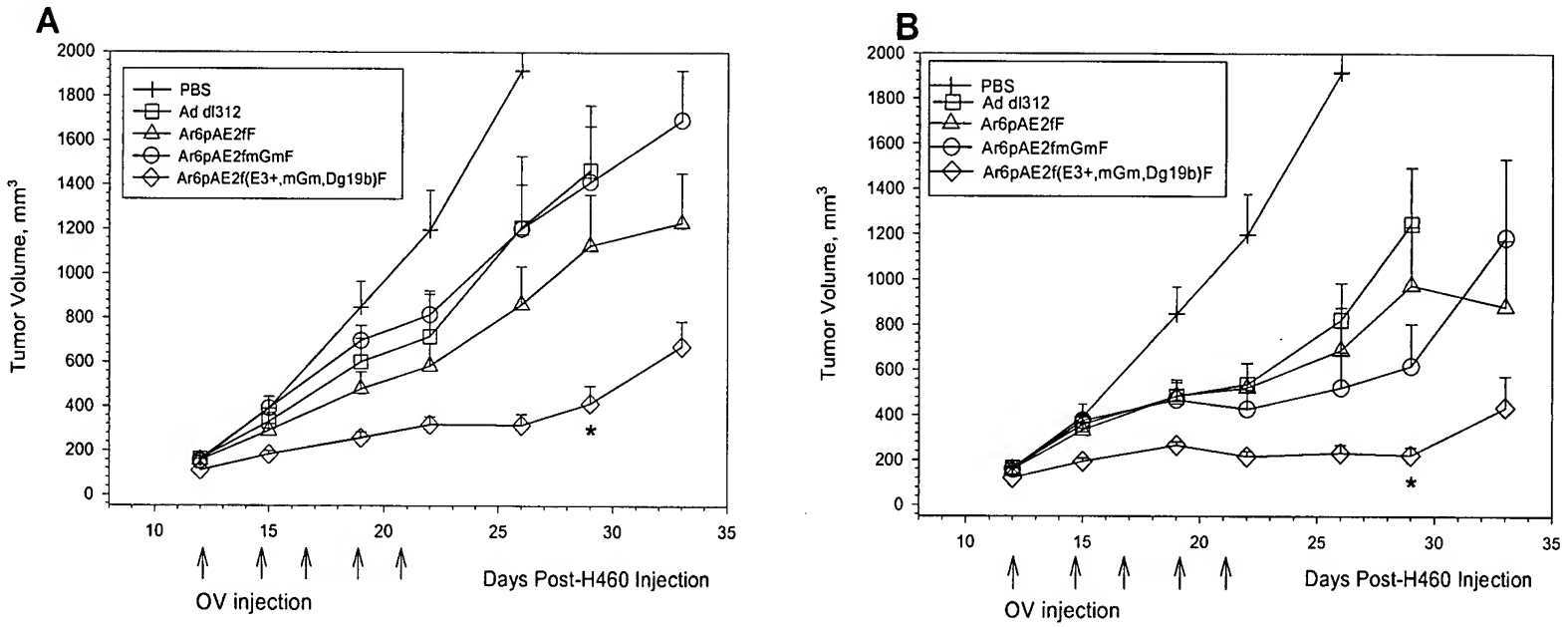
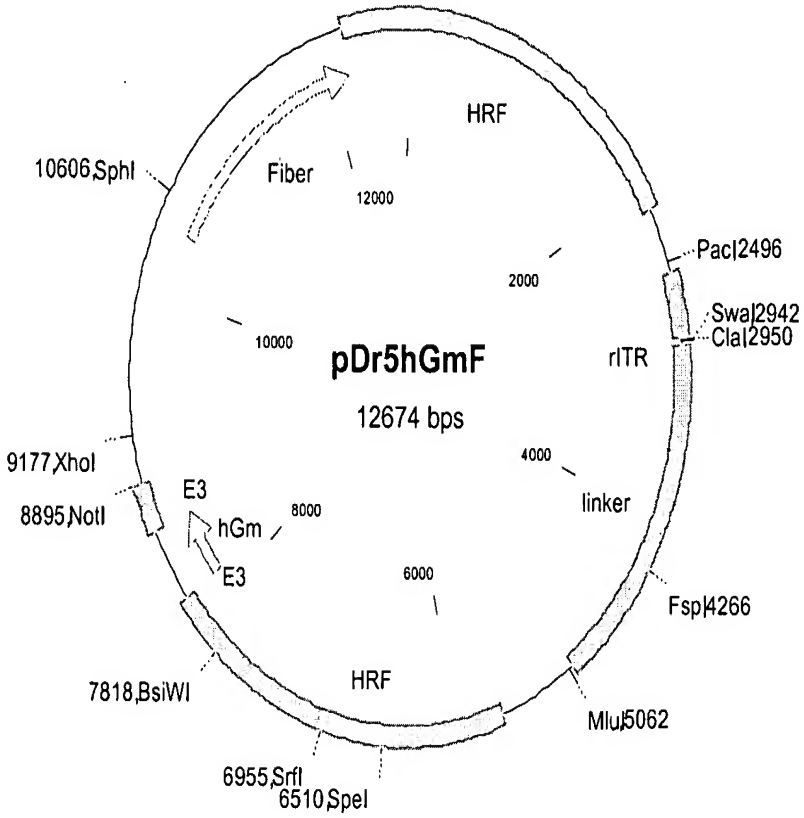


Figure 32. Schematic diagram of adenovirus pDr5hGmF and pDr5mGmF right donor plasmids.

A. pDr5hGmF



B. pDr5mGmF

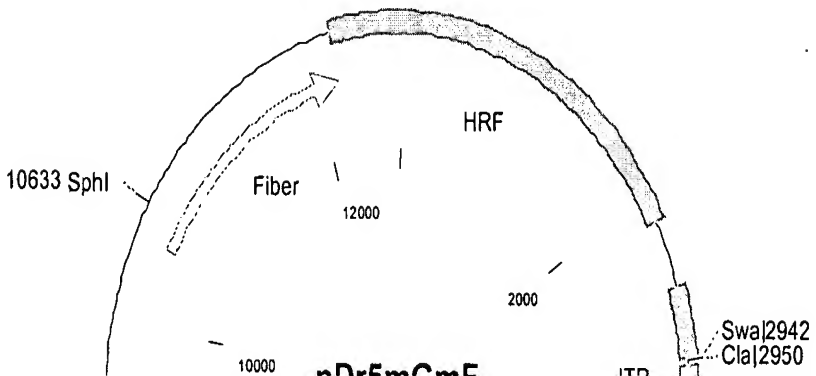


Figure 33. Pathway used to generate the pAr15pAE2fhGmF plasmid.

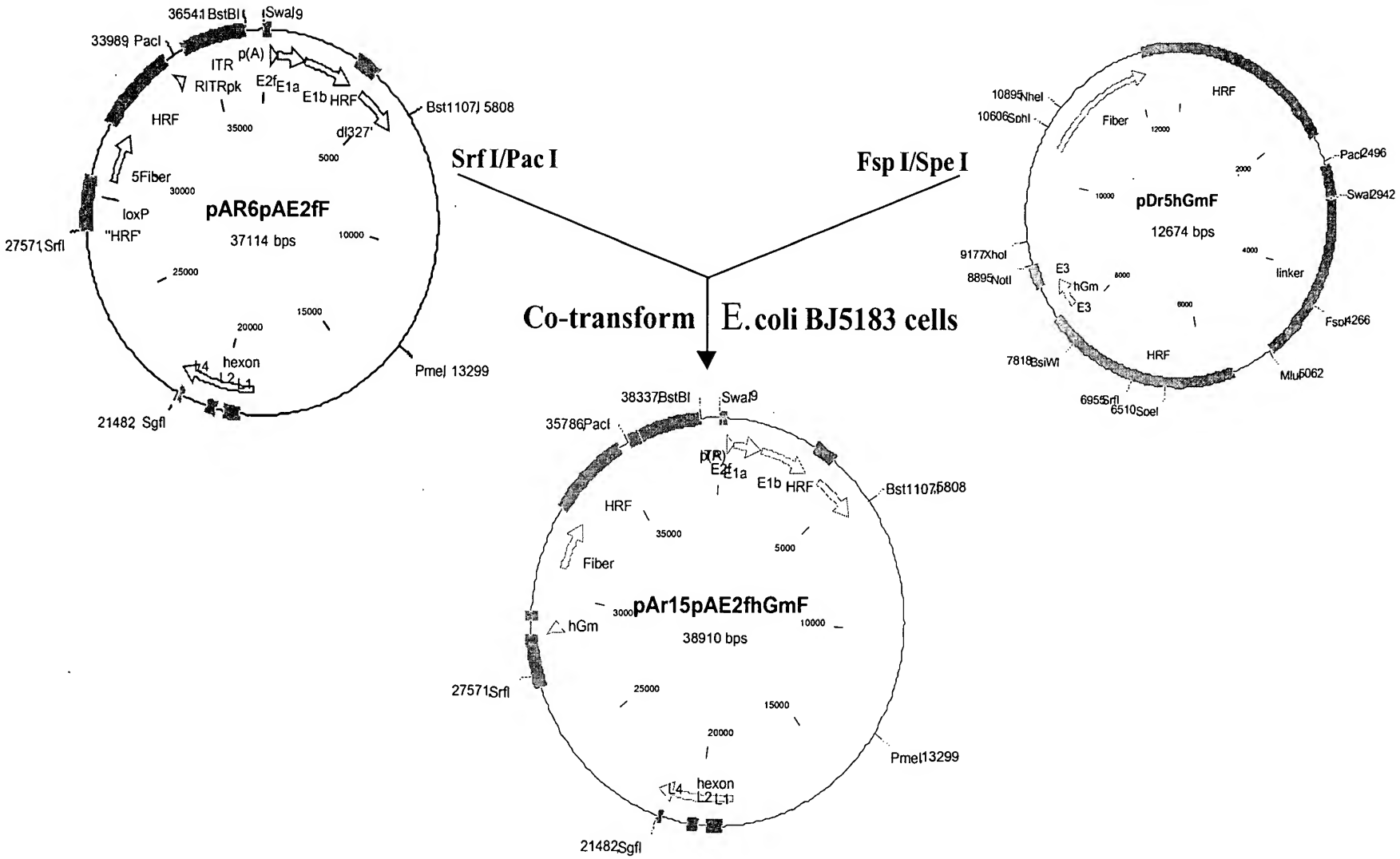


Figure 34. Pathway used to generate the pAr15pAE2fmGmF plasmid.

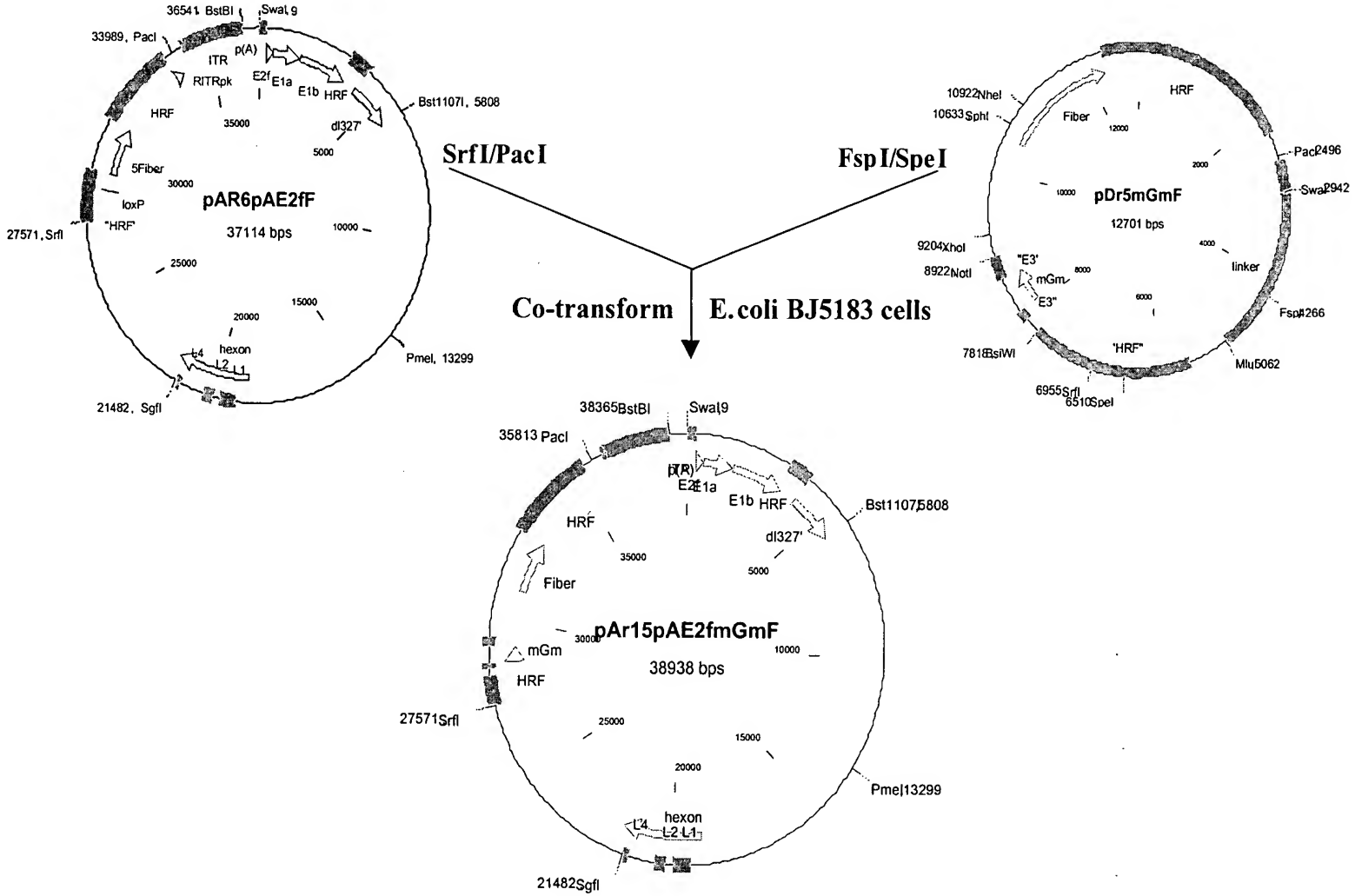
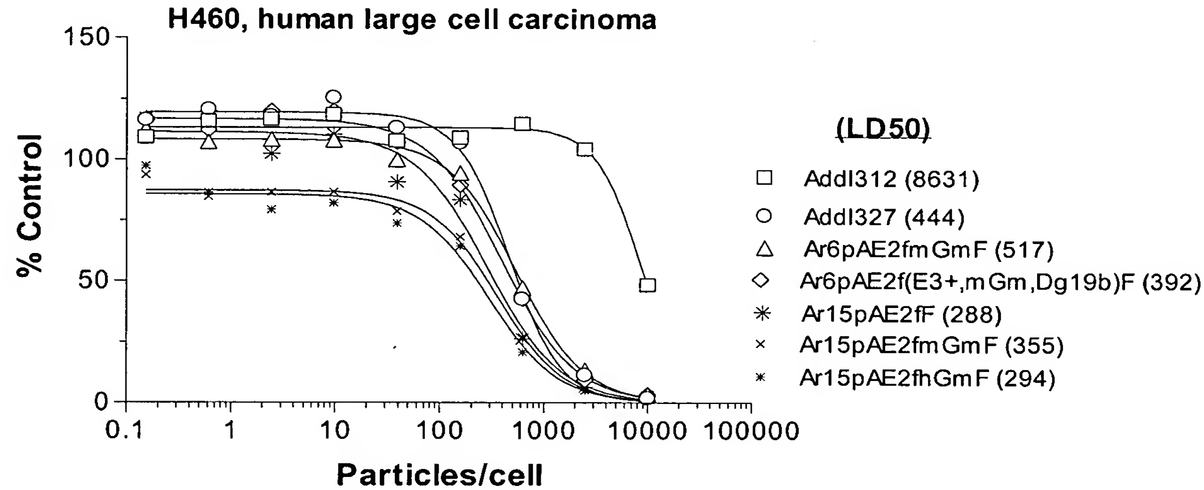


Figure 35. MTS assay of Ar15pAE2fhGmF and Ar15pAE2fmGmF vectors on H460 and Hep3B tumor cell lines.



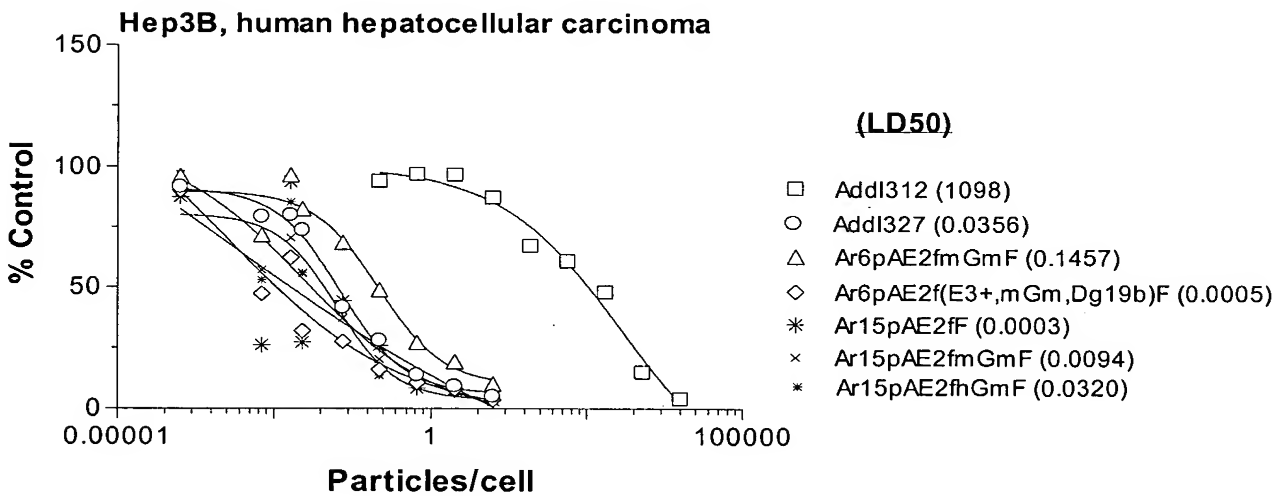


Figure 36. GM-CSF expression mediated by Ar15pAE2fhGmF and Ar15pAE2fmGmF vectors in infected H460 cells detected by ELISA.

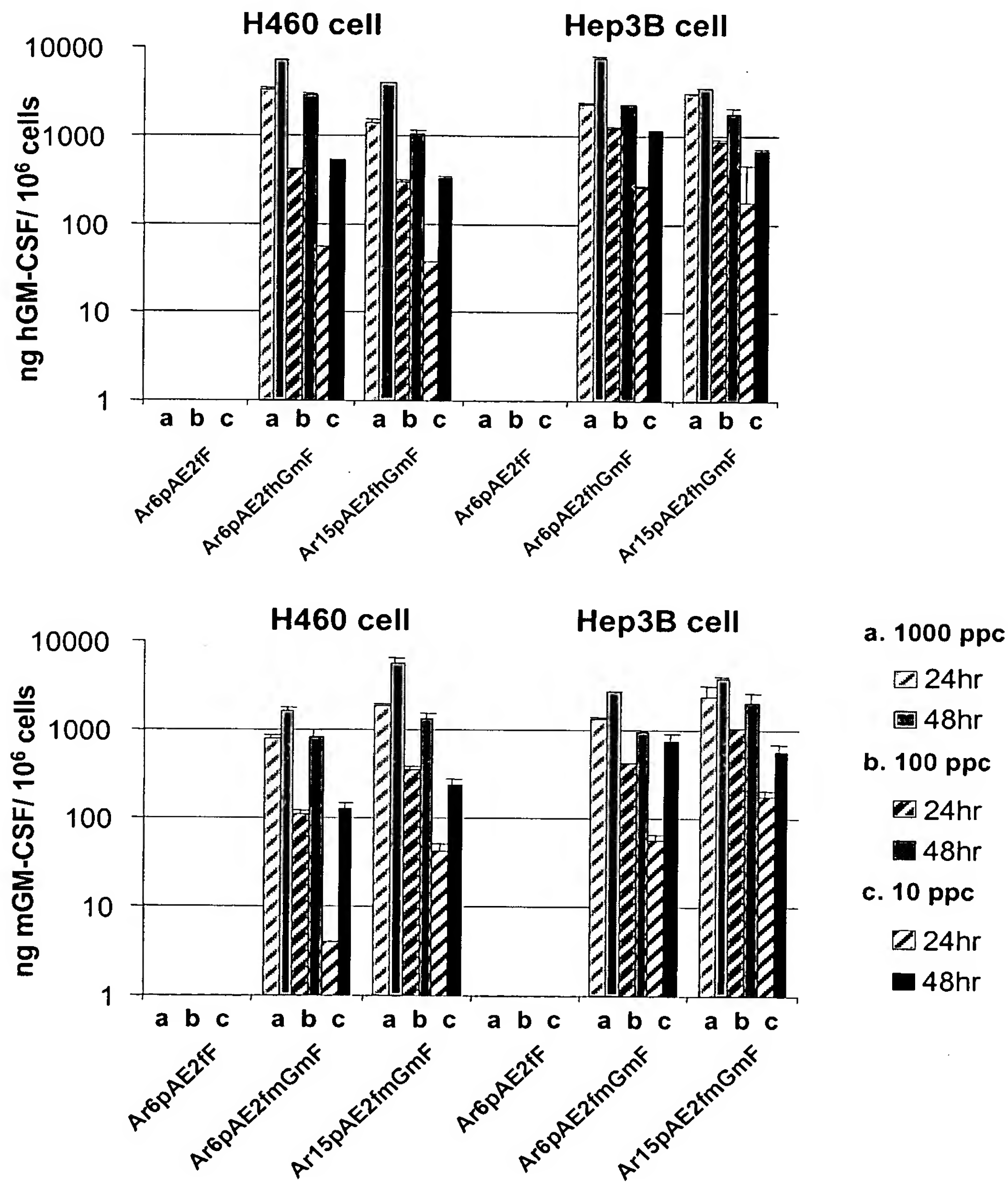


Figure 37. Schematic Diagram of PCR and Overlap PCR for ΔE3-14.7 plasmids

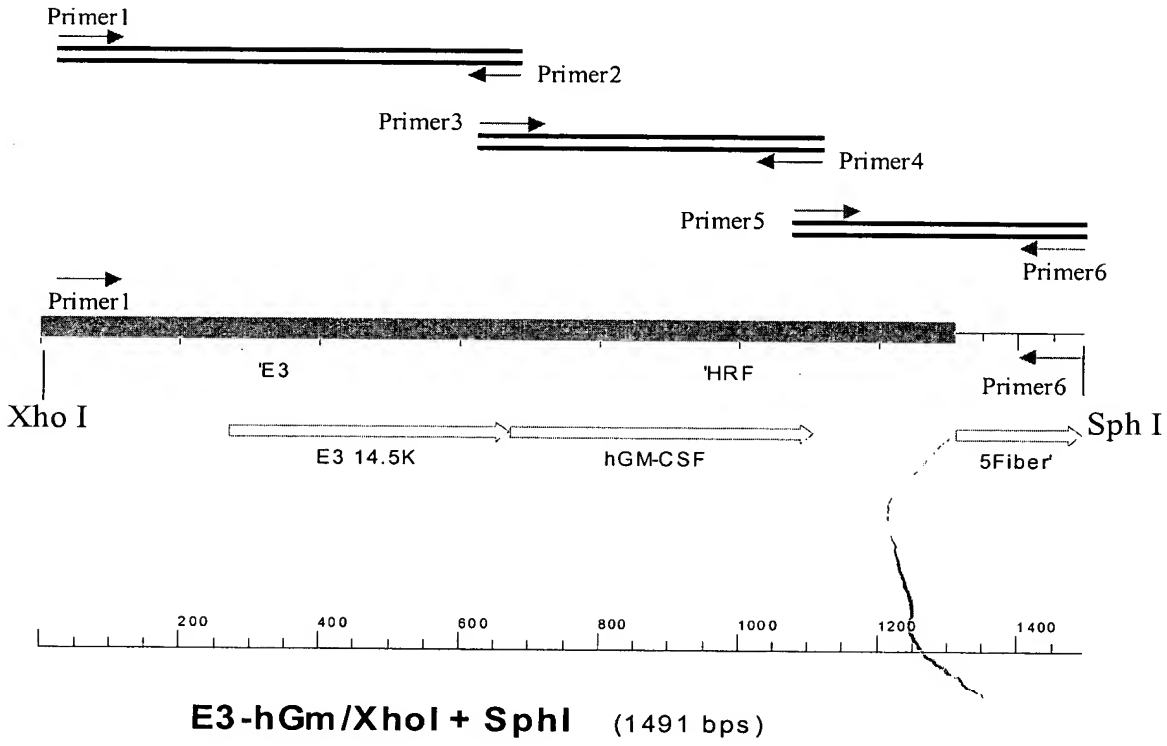


Figure 38. Schematic Diagram of ΔE3-14.7 Vectors

a. Sequence of native E3-14.5/E3-14.7 junction:

ggaggagATGacTGAttaggtac (Seq ID NO:14)

E3 14.5kDa--------G G D D STOP (Seq ID NO:16)

M------E3 14.7kDa

b. Sequence of the Ar16pAE2fhGm vector at the junction engineered between the E3-14.5 gene and human GM-CSF cDNA:

ggaggagacgacTGACC atg......... (Seq ID NO:15)

E3 14.5kDa-------G G D D STOP M------GMCSF

Figure 39. Pathway Used to Generate the pAr16pAE2fhGmF Large Plasmid

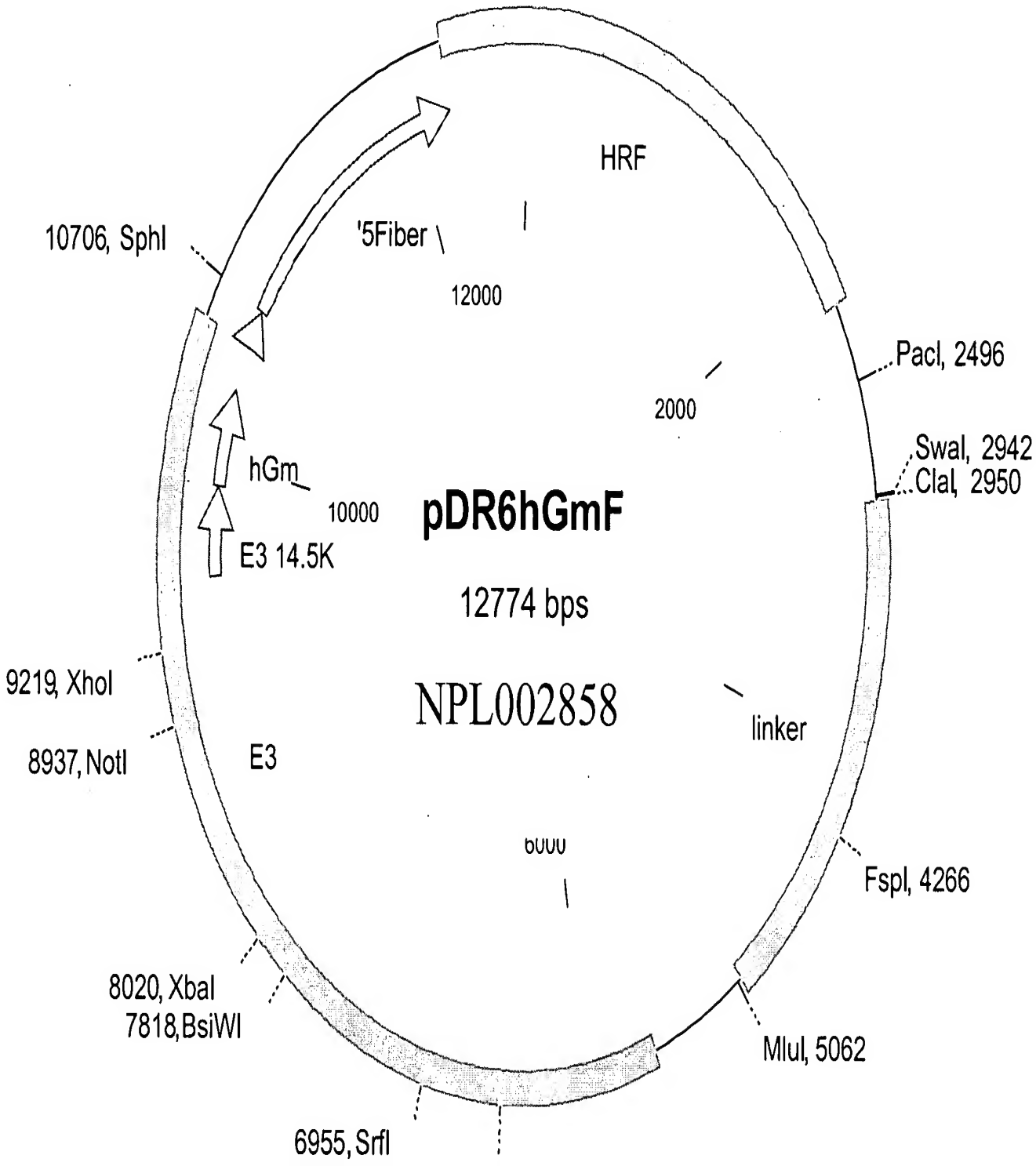


Figure 40. Pathway Used to Generate the pAr16pAE2fhGmF Large Plasmid

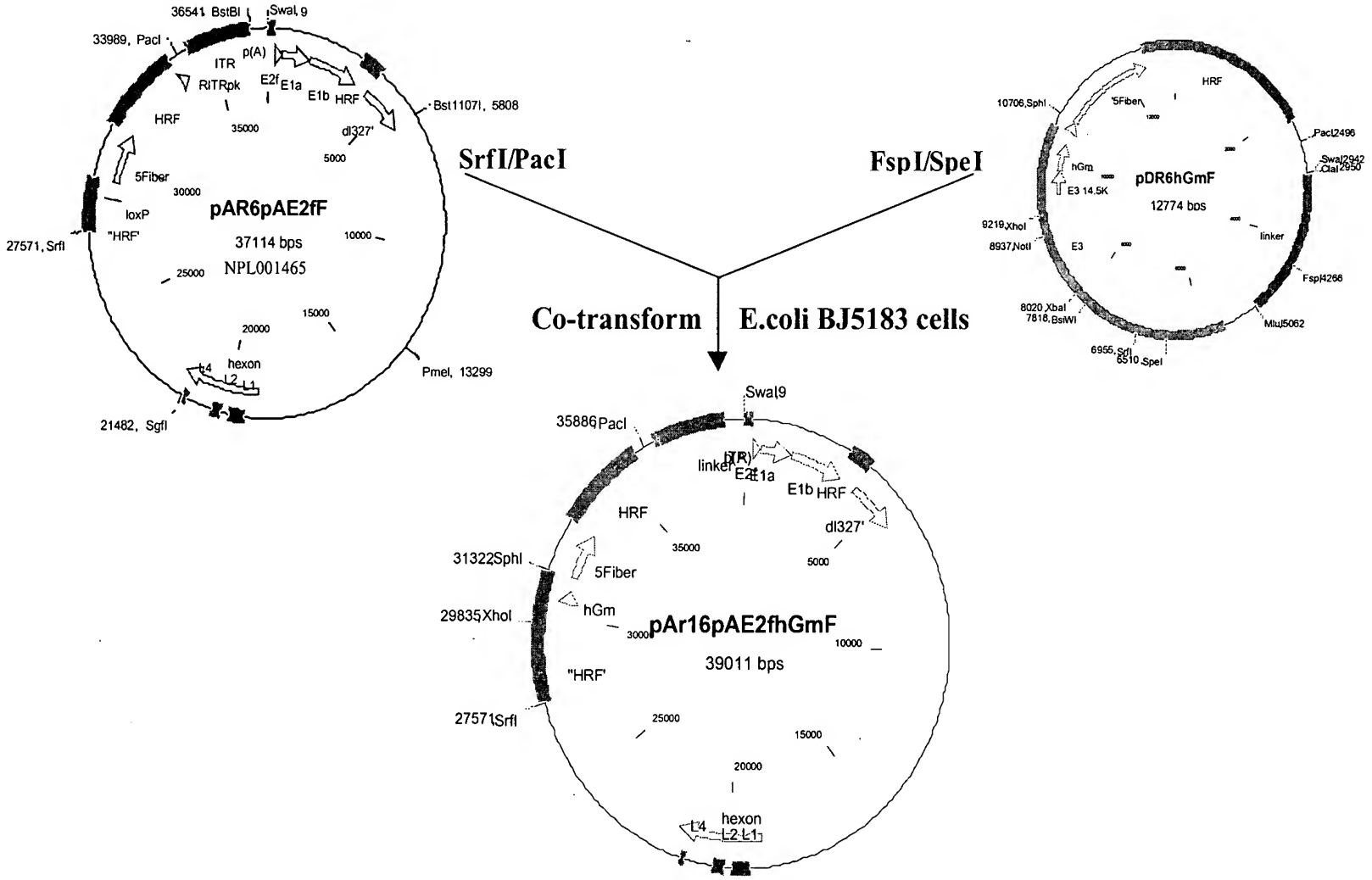


Figure 41. MTS Assay of ΔE3-14.7 hGM-CSF Vector on H460 Tumor Cell Line

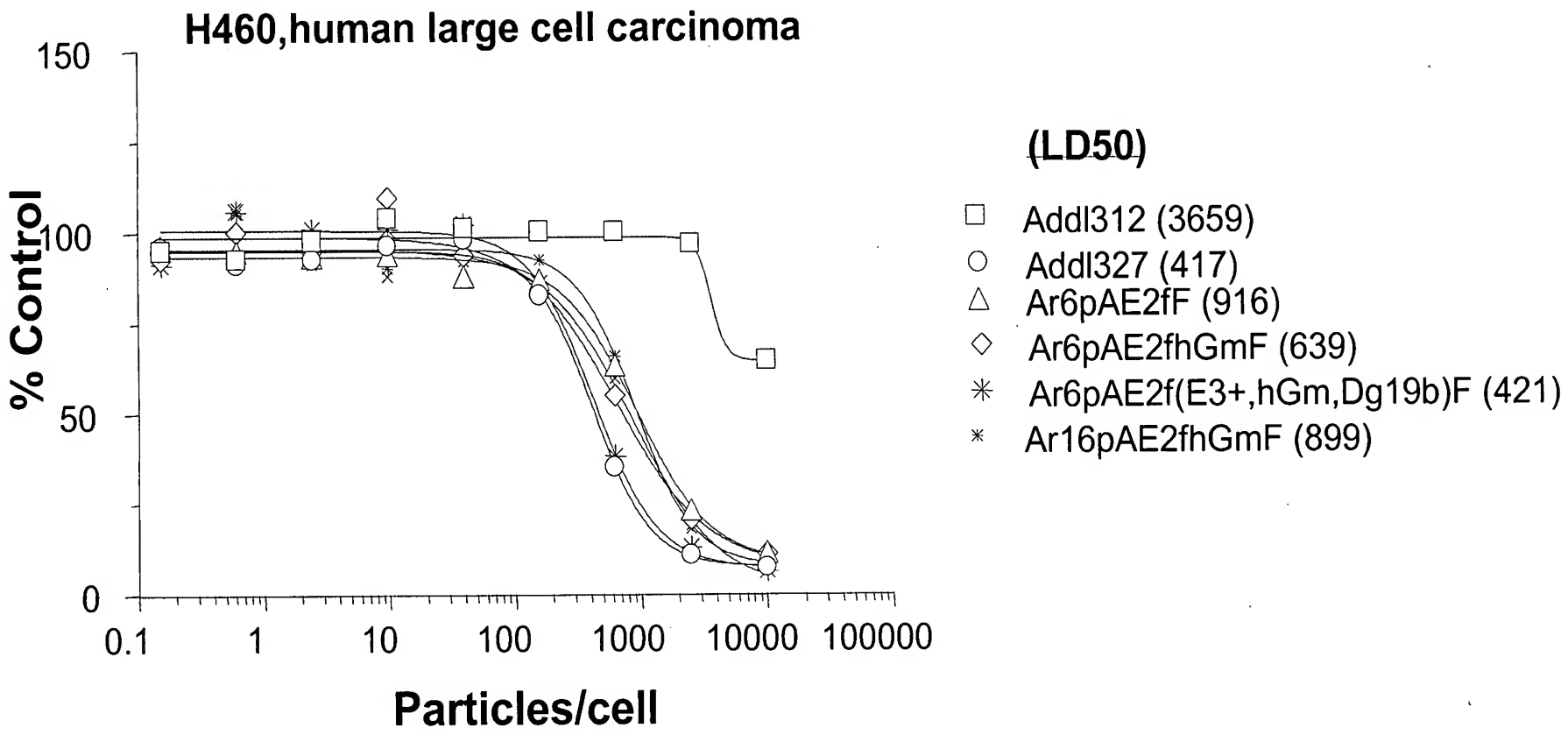


Figure 42. GM-CSF Expression Mediated by ΔE3-14.7 hGM-CSF Vector (Ar16pAE2fhGmF) compared to Ar6pAE2fF, Ar6pAE2fhGmF and Ar6pAE2f(E3+,hGm,Dg19)F in Infected H460 Cells 24 Hours Post-Infection

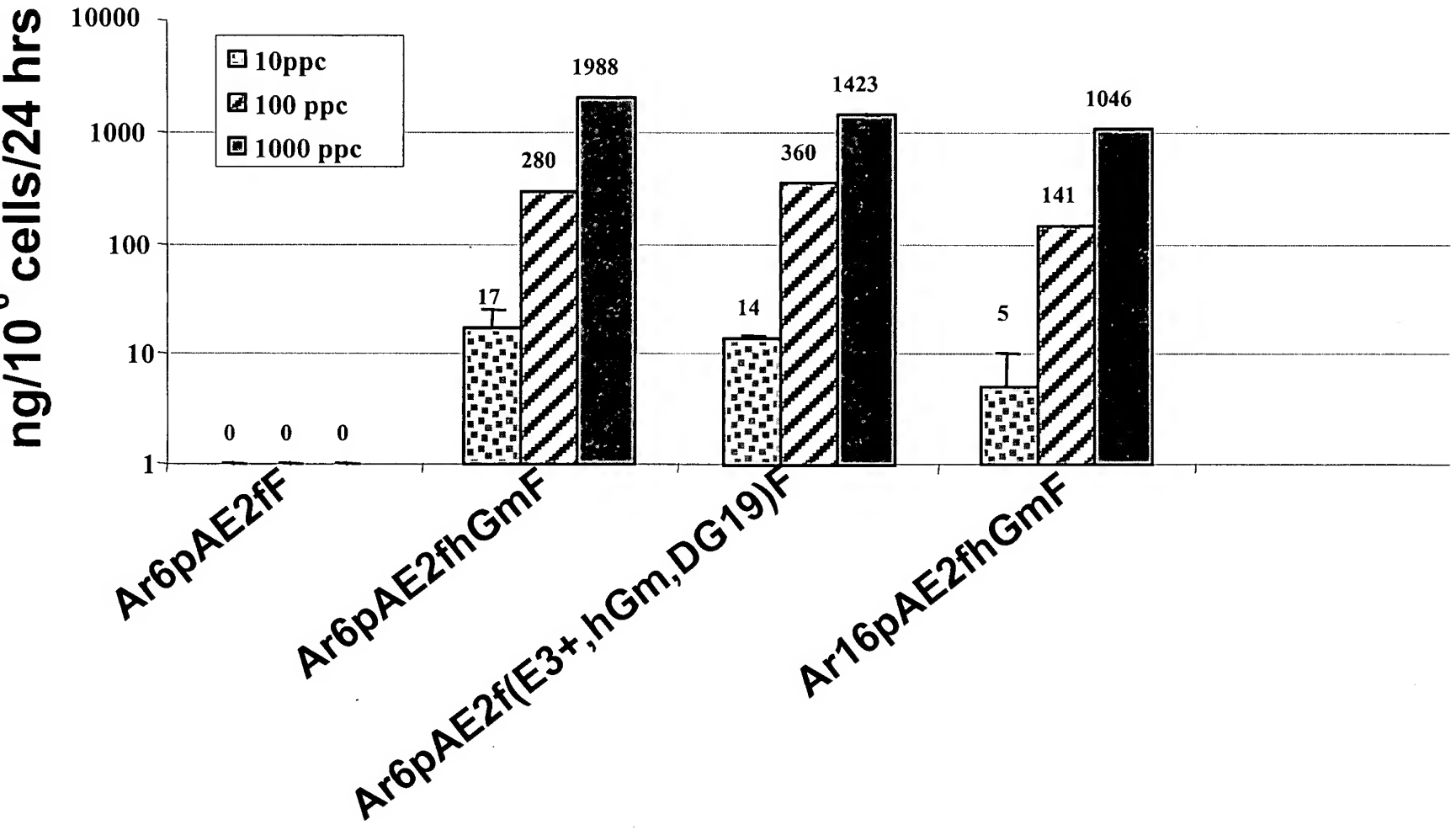
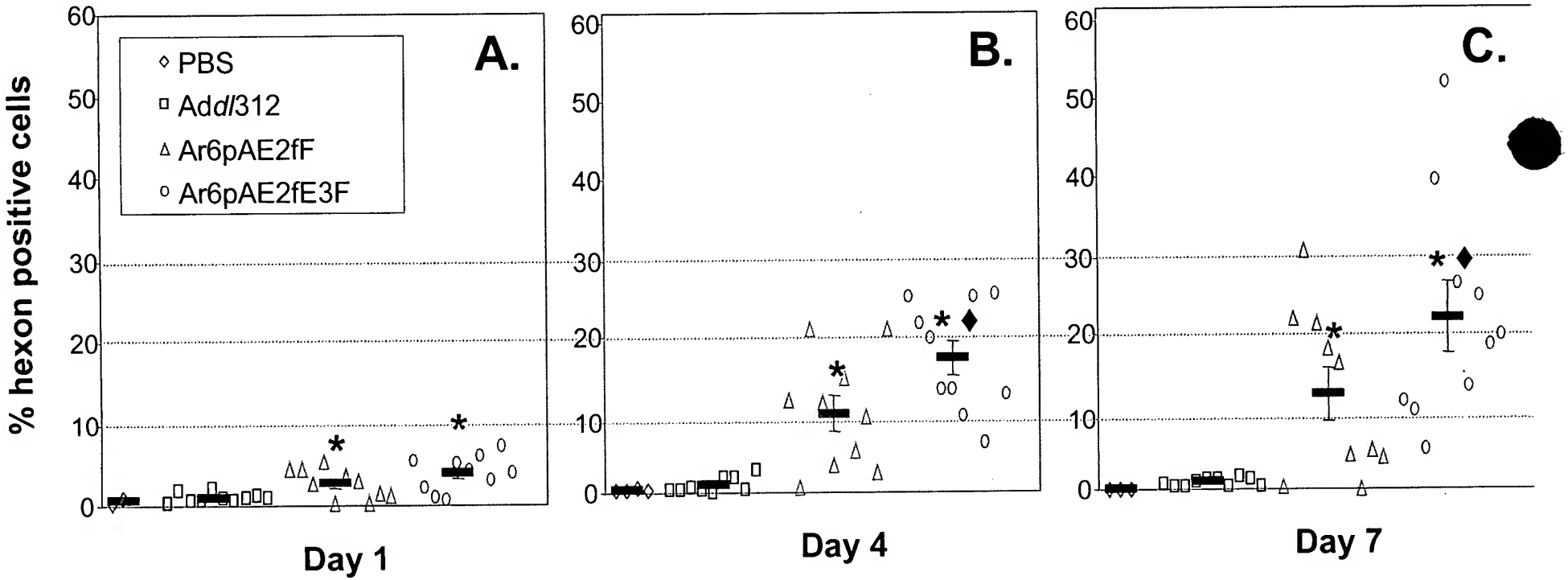
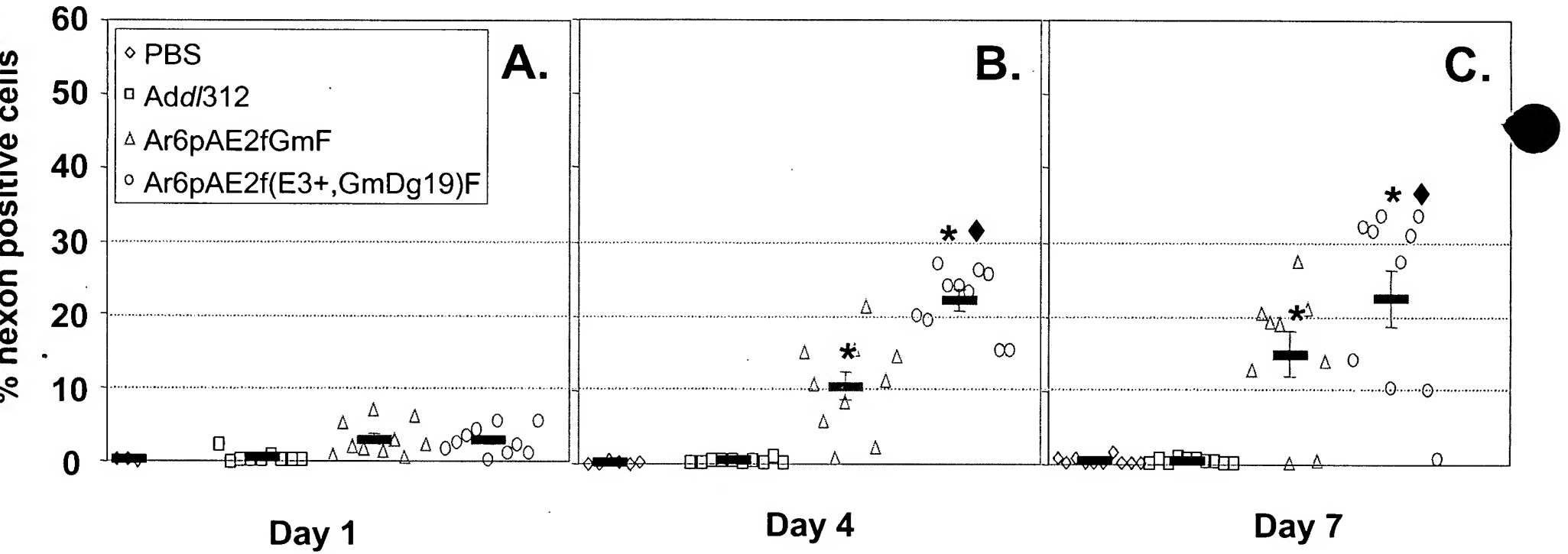


Figure 43. Spread of adenoviruses in H460 xenograft tumors detected by FACS.



*: p<0.05 between Ar6pAE2fF or Ar6pAE2fE3F and Add/312, ANOVA
•: p<0.05 between Ar6pAE2fF and Ar6pAE2fE3F vectors, ANOVA

igure 44. Spread of adenoviruses in Hep3B xenograft tumors detected by FACS.



p<0.05 between Ar6pAE2fhGmF or Ar6pAE2f(E3+,hGm,Dg19)F and Add/312, ANOVA
p<0.05 between Ar6pAE2fhGmF and Ar6pAE2f(E3+,hGm,Dg19)F vectors, ANOVA

Figure 45: Flowchart for construction of pAR17pAE2fFTrtex:

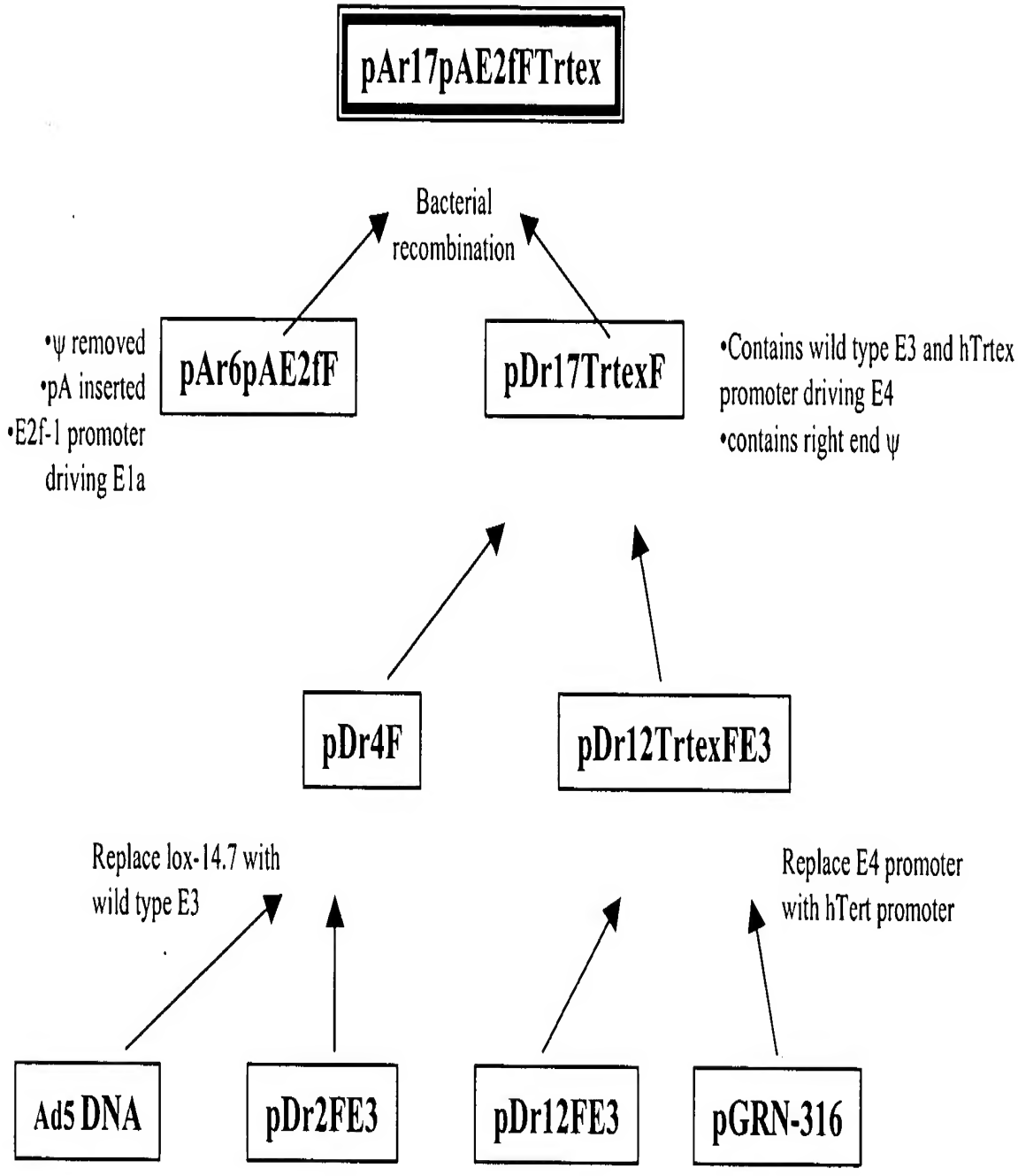
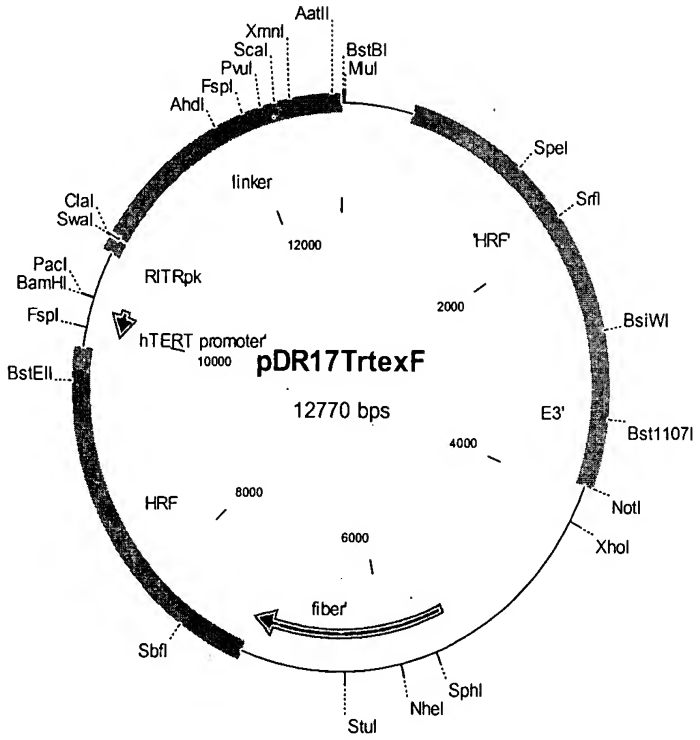


Figure 46: Plasmids used to create oncolytic vector Ar17pAE2fFTrtex



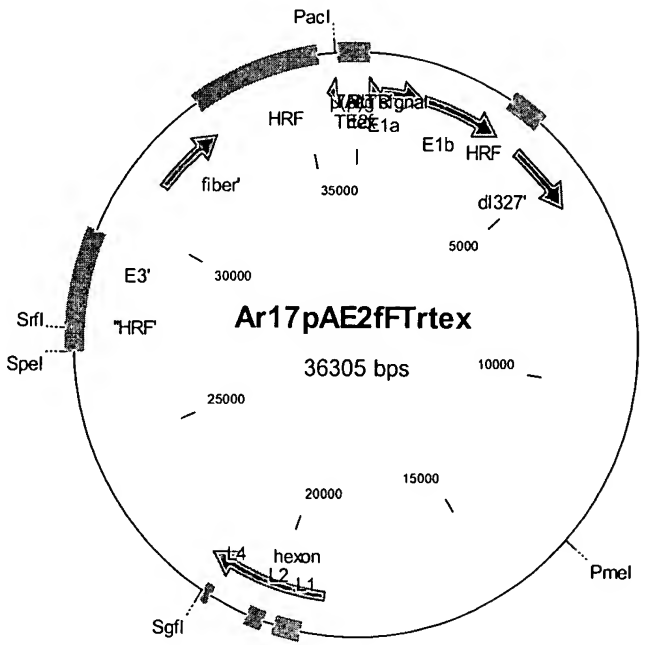


Figure 47: Sequence of the right end of Ar17pAE2fFTrtex (Seq ID NO:17).

```
35351  agtgctaaaa agcgaccgaa atagcccggg ggaatacata cccgcaggcg
35401  tagagacaac attacagccc ccataggagg tataacaaaa ttaataggag
35451  agaaaaacac ataaacacct gaaaaaccct cctgcctagg caaaatagca
35501  ccctcccgct ccagaacaac atacagcgct tcacagcggc agcctaacag
35551  tcagccttac cagtaaaaaa gaaaacctat taaaaaaaca ccactcggat
35601  caattcgcgg gggtggccgg ggccagggct tcccacgtgc gcagcaggac
35651  gcagcgctgc ctgaaactcg cgccgcgagg agagggcggg gccgcggaaa
35701  ggaaggggag gggctgggag ggcccggagg gggctgggcc ggggacccgg
35751  gaggggtcgg gacggggcgg ggtccgcgcg gaggaggcgg agctggaagg
35801  tgaaggggca ggacgggtgc ccgggtcccc agtccctccg ccacgtgggg
35851  ctaggatcct taattaagaa ttctacaatt cccaacacat acaagttact
35901  ccgccctaaa accctgggcg agtctccacg taaacggtca aagtccccgc
35951  ggccctagac aaatattacg cgctatgagt aacacaaaat tattcagatt
36001  tcacttcctc ttattcagtt ttcccgcgaa aatggccaaa tcttactcgg
36051  ttacgcccaa atttactaca acatccgcct aaaaccgcgc gaaaattgtc
36101  acttcctgtg tacaccggcg cacaccaaaa acgtcacttt tgccacatcc
36151  gtcgcttaca tgtgttccgc cacacttgca acatcacact tccgccacac
36201  tactacgtca cccgccccgt tcccacgccc cgcgccacgt cacaaactcc
36251  acccccctcat tatcatattg gcttcaatcc aaaataaggt atattattga
36301  tgatg
```

Figure 48: Diagram of Ar17pAE2fFTrtex.

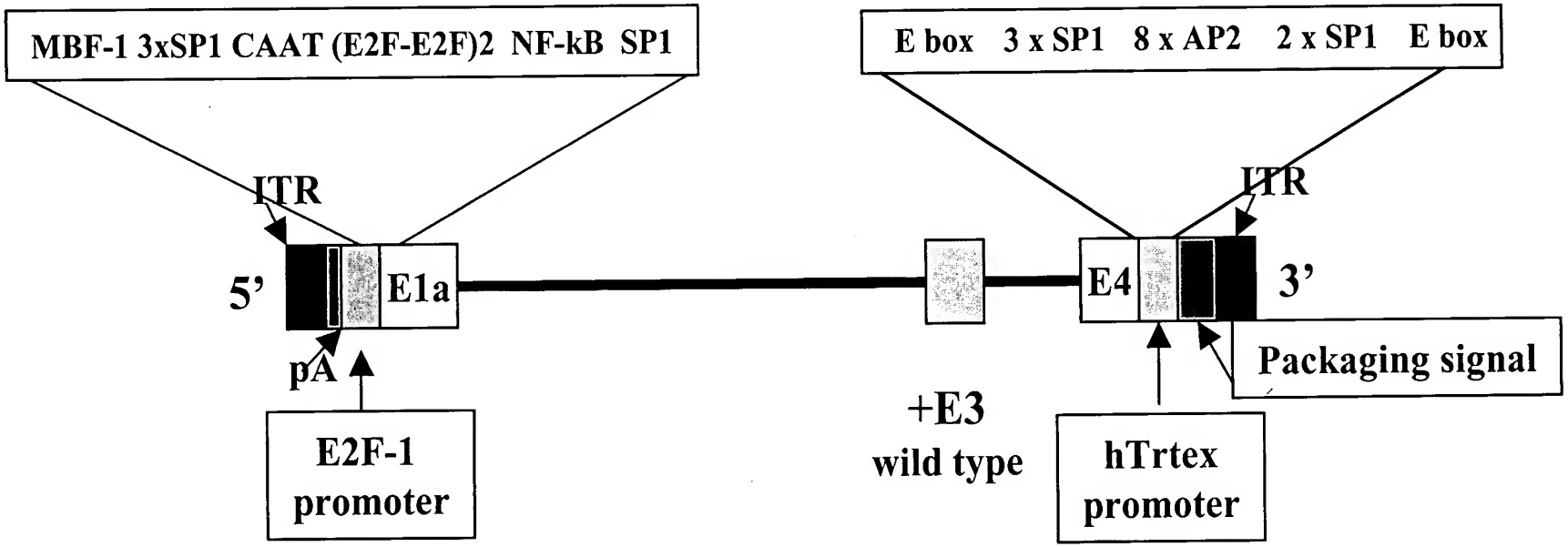


Figure 49. E4 expression is dependent on the hTERT promoter

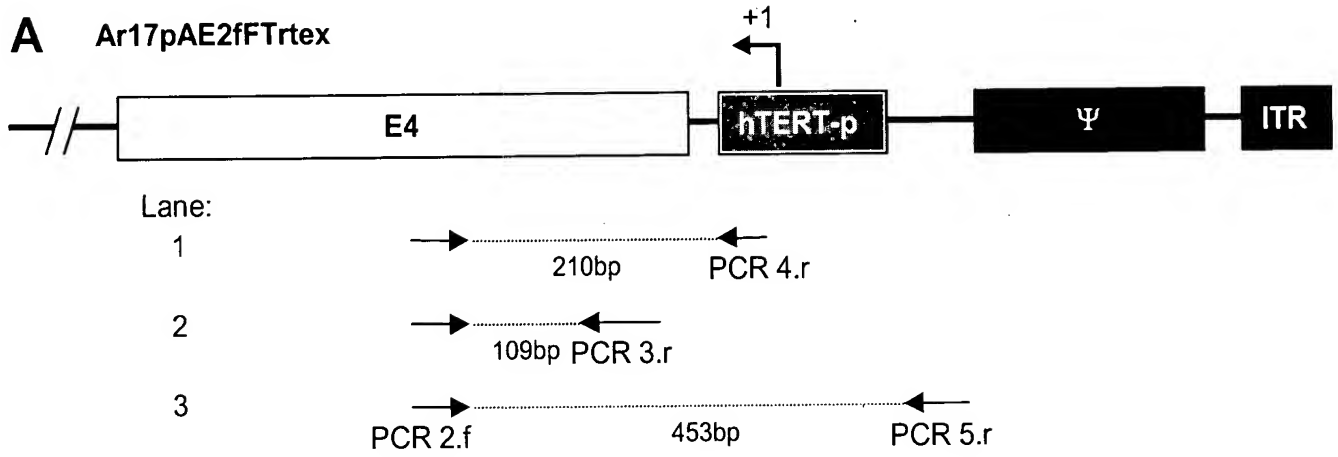


Figure 50. E4 transcription start sites in Ar17pAE2fFTrtex (Seq ID NO:21)

35521 ATACAGCGCT TCACAGCGGC AGCCTAACAG TCAGCCTTAC CAGTAAAAAA GAAAACCTAT

ExtP1 ←

35581 TAAAAAAACA CCACTCGGAT CAATTCGCGG GGGTGGCCGG GGCCAGGGCT TCCCACGTGC

← ← ←

35641 GCAGCAGGAC GCAGCGCTGC CTGAAACTCG CGCCGCGAGG AGAGGGCGGG GCCGCGGAAA

←

35701 AGGAACGGGA CGGGCTGGGA TGGCCCGGAA GGGGCTGGGC CGGGGACCCG GGAAGGGTTC

35761 GGGACGGGGC GGGGTTCCGC GCGGACGAGG CGGAGCTGGA AGGTGAAGGG GCAGGACCGG

35821 TGCCCGGGTC CCCAGTCCCT CCGCCACGTG GGGCTAGGAT CCTTAATTAA GAATTCTACA

35881 ATTCCCAACA CATACAAGTT ACTCCGCCCT AAAACCCTGG GCG

Figure 51. Efficacy of Ar17pAE2fFTrtex in Hep3B model.

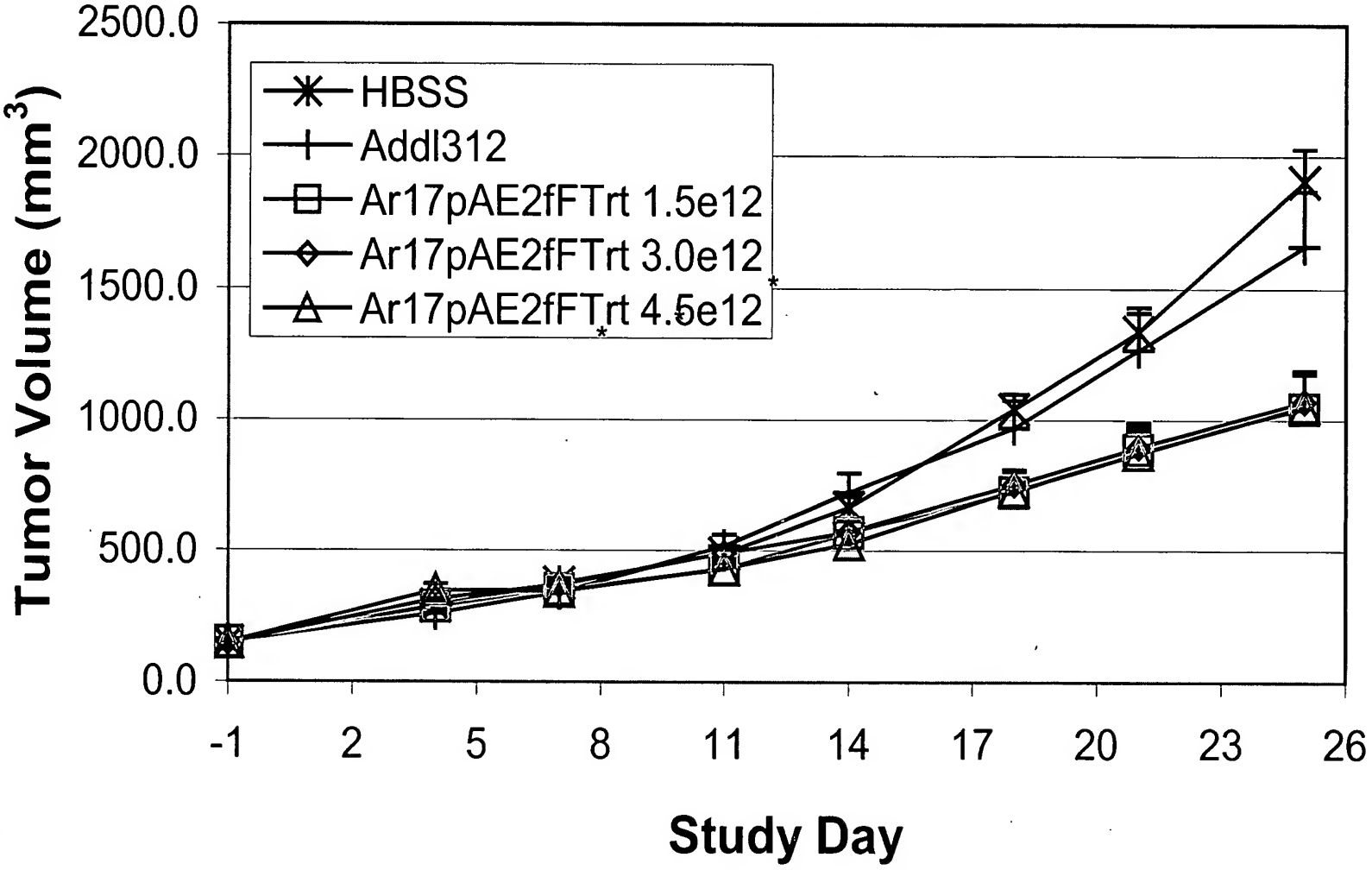


Figure 52. Effect of Ar17pAE2fFTrtex on survival.

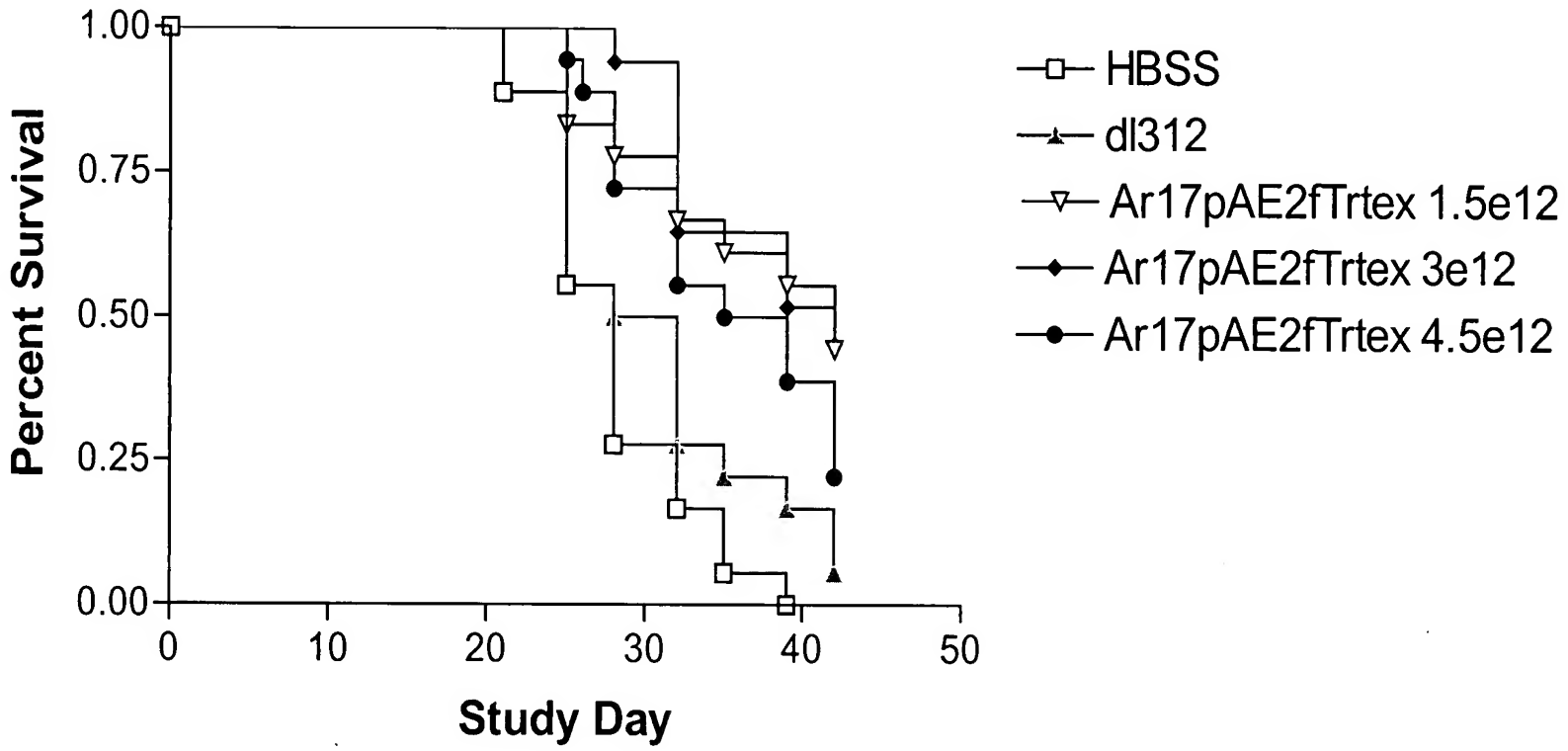


Figure 53. Body weight changes

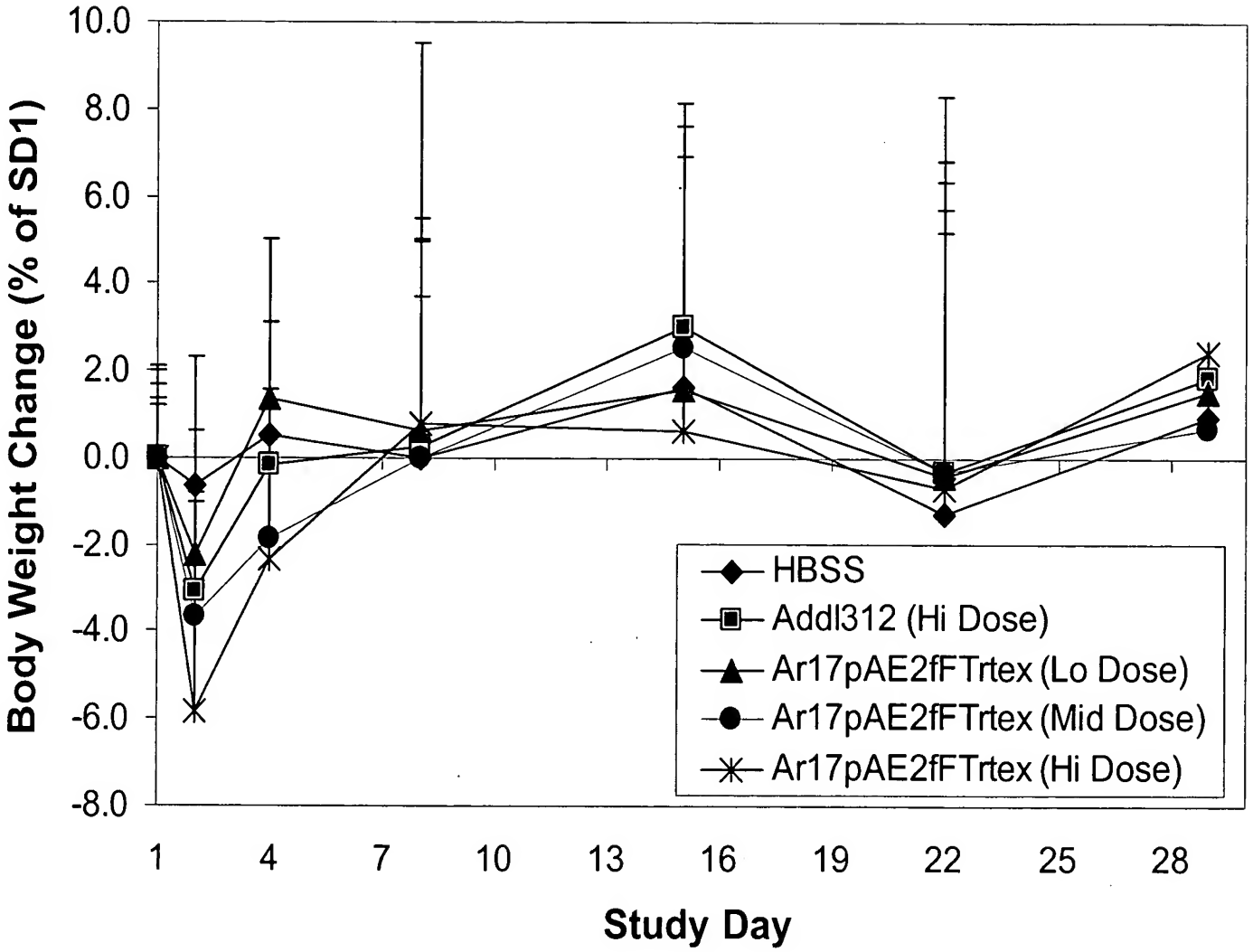


Figure 54. Efficacy of Ar17pAE2fFTrtex in Hep3B model.

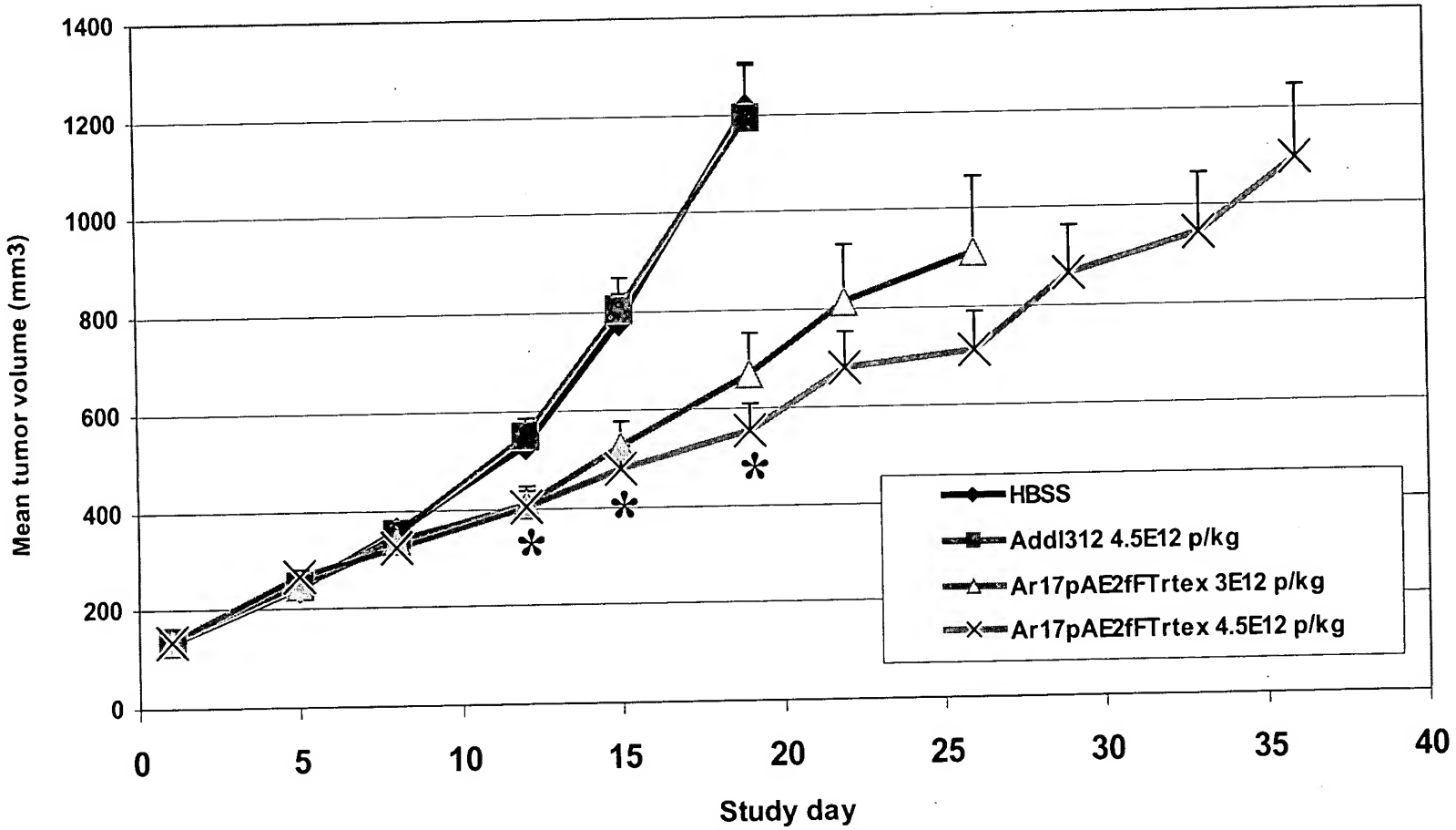


Figure 55. Effect of Ar17pAE2fFTrtex on survival.

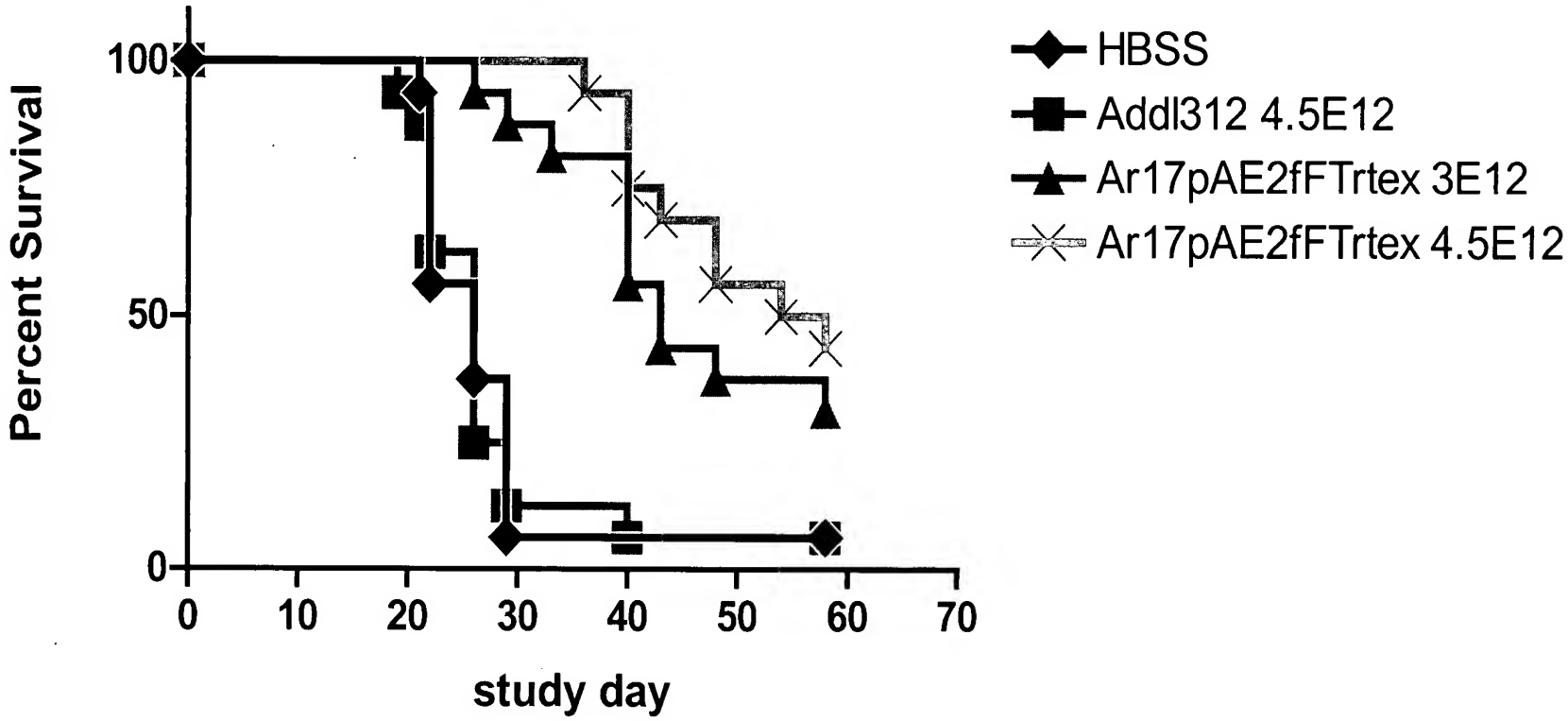


Figure 56. Body weight changes

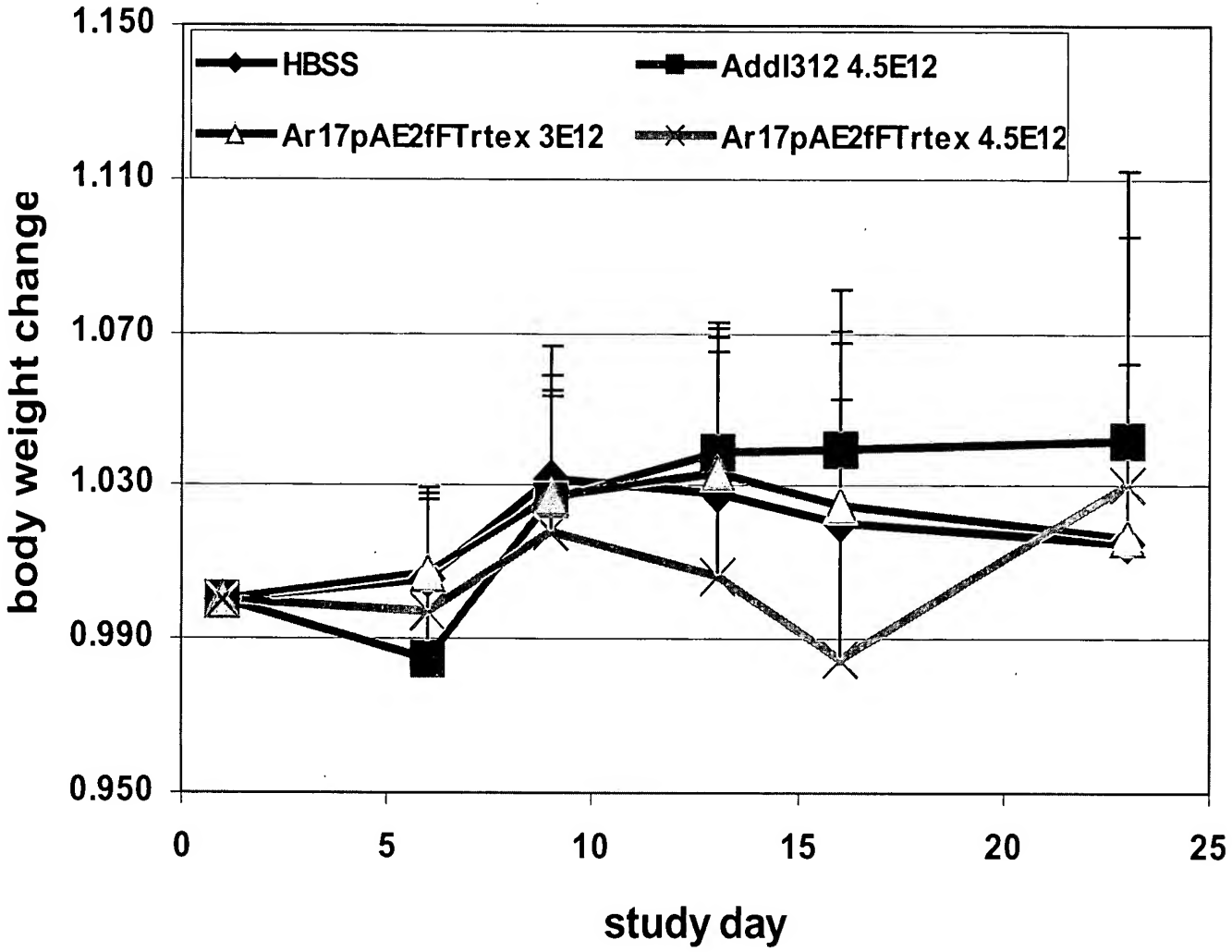


Figure 57 Dose-dependent anti-tumor efficacy

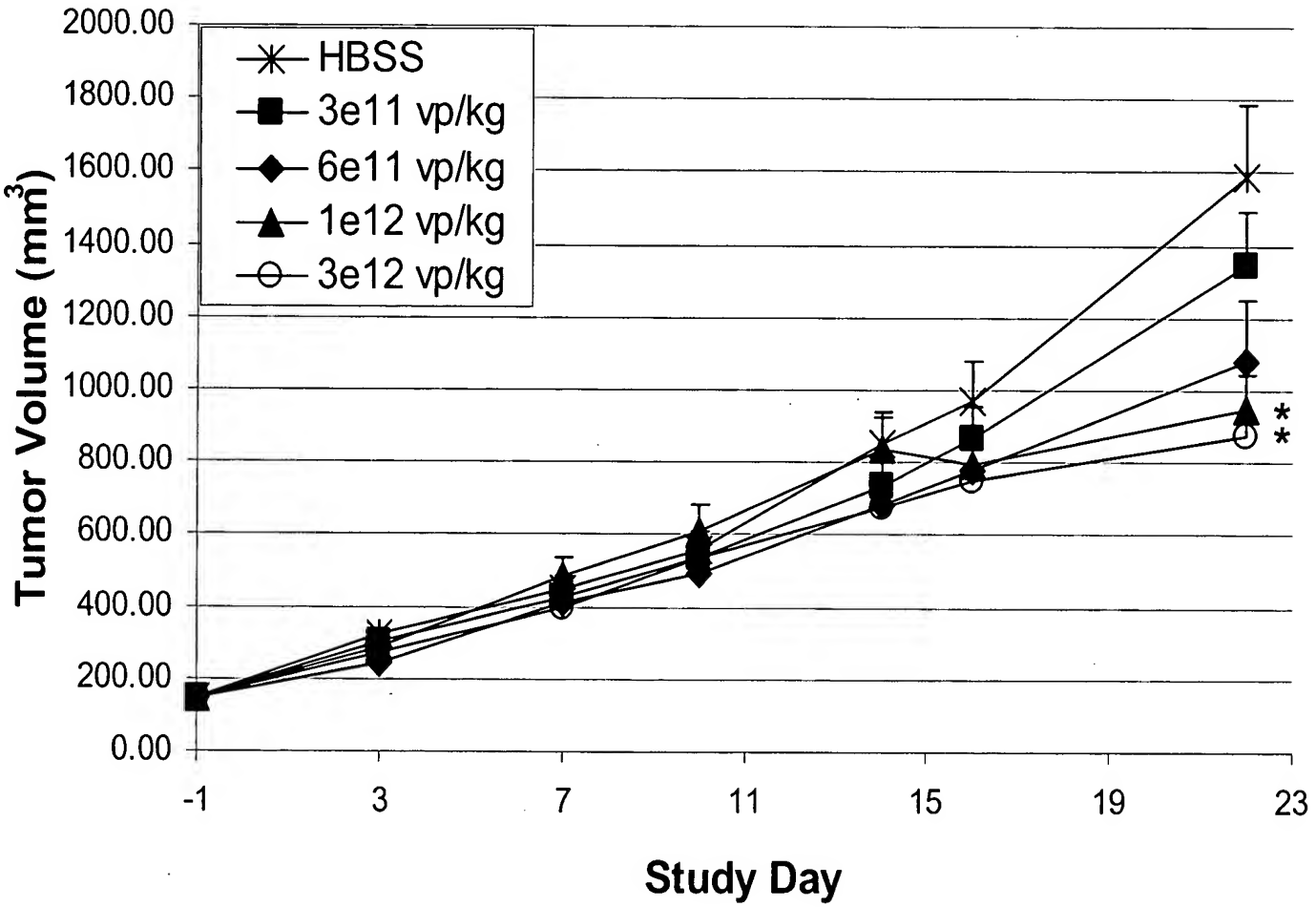


Figure 58. Individual tumor volumes following intravenous administration of Ar17pAE2fFTrtex

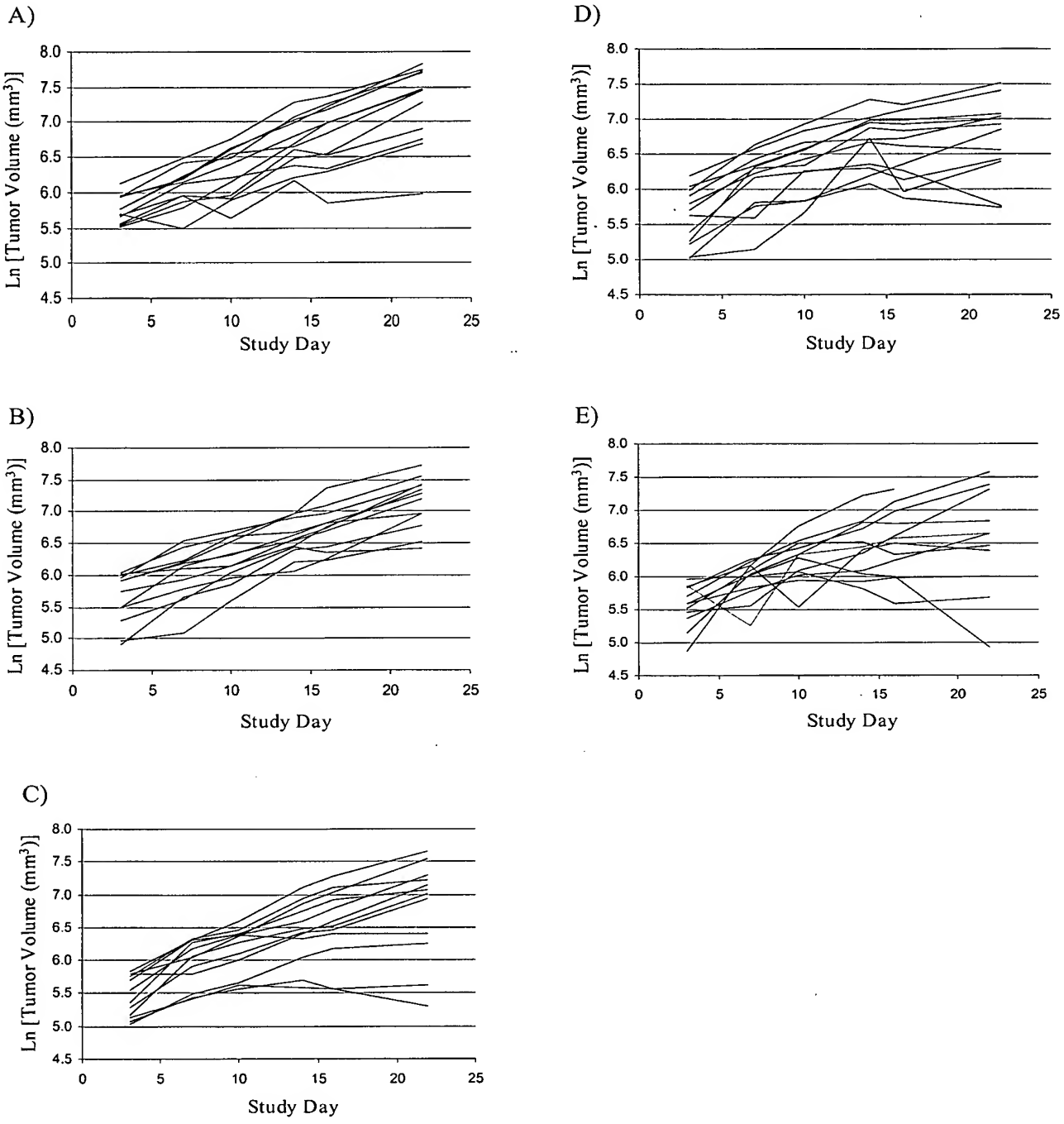


Figure 59. Effect of Ar17pAE2fFTrtex on survival.

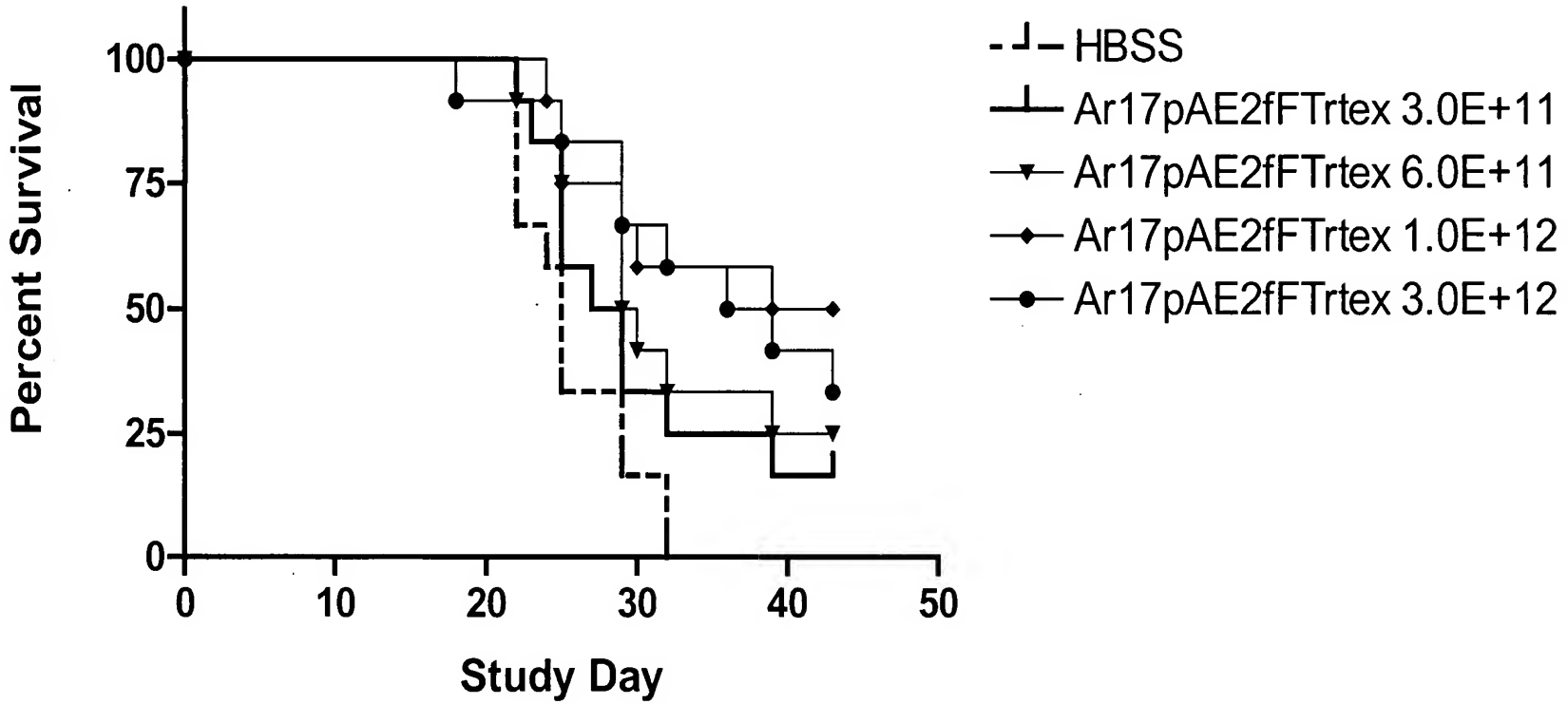


Figure 60. Body weight (% change)

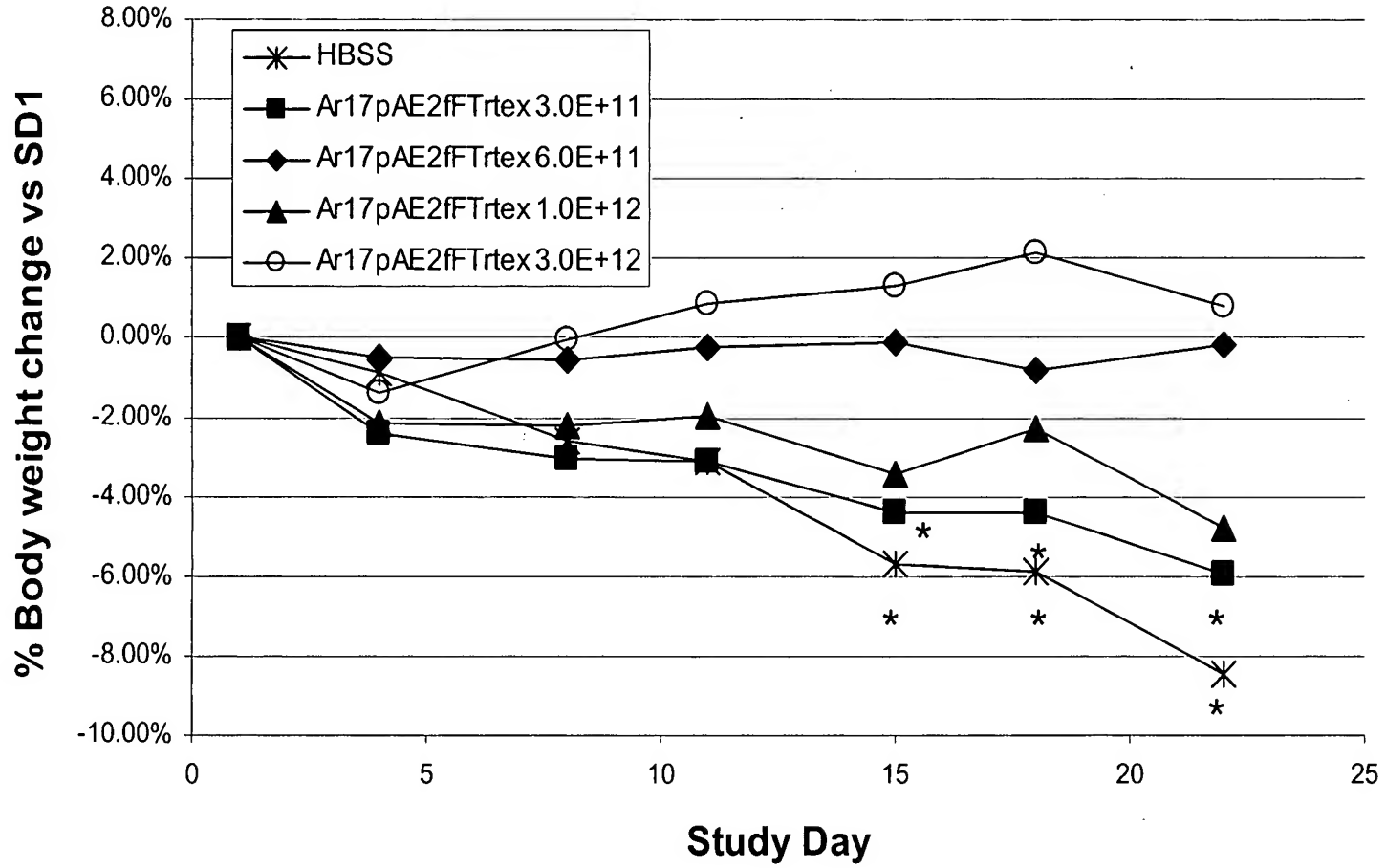


Figure 61.

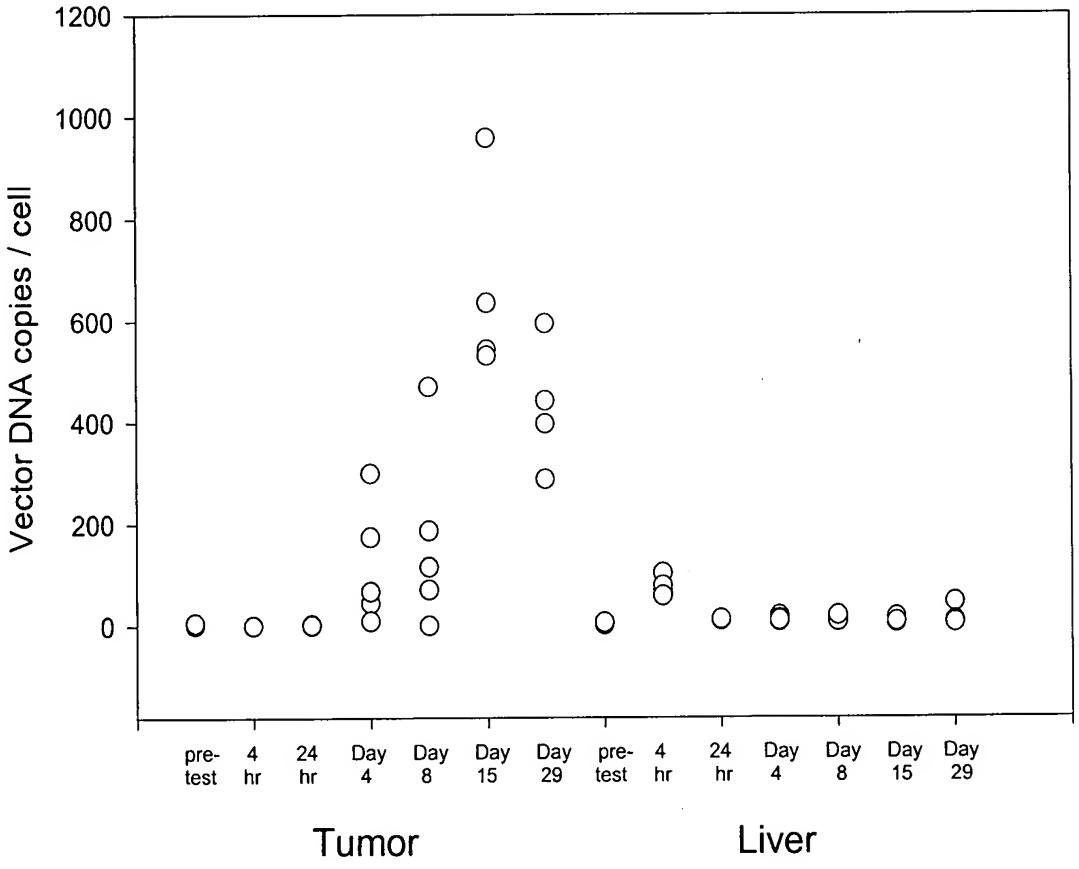

Vector DNA copies / cell
1200
1000
800
600
400
200
0
pre-test
4 hr
24 hr
Day 4
Day 8
Day 15
Day 29
pre-test
4 hr
24 hr
Day 4
Day 8
Day 15
Day 29
Tumor
Liver

Figure 62. Effect on body weight in SCID mice

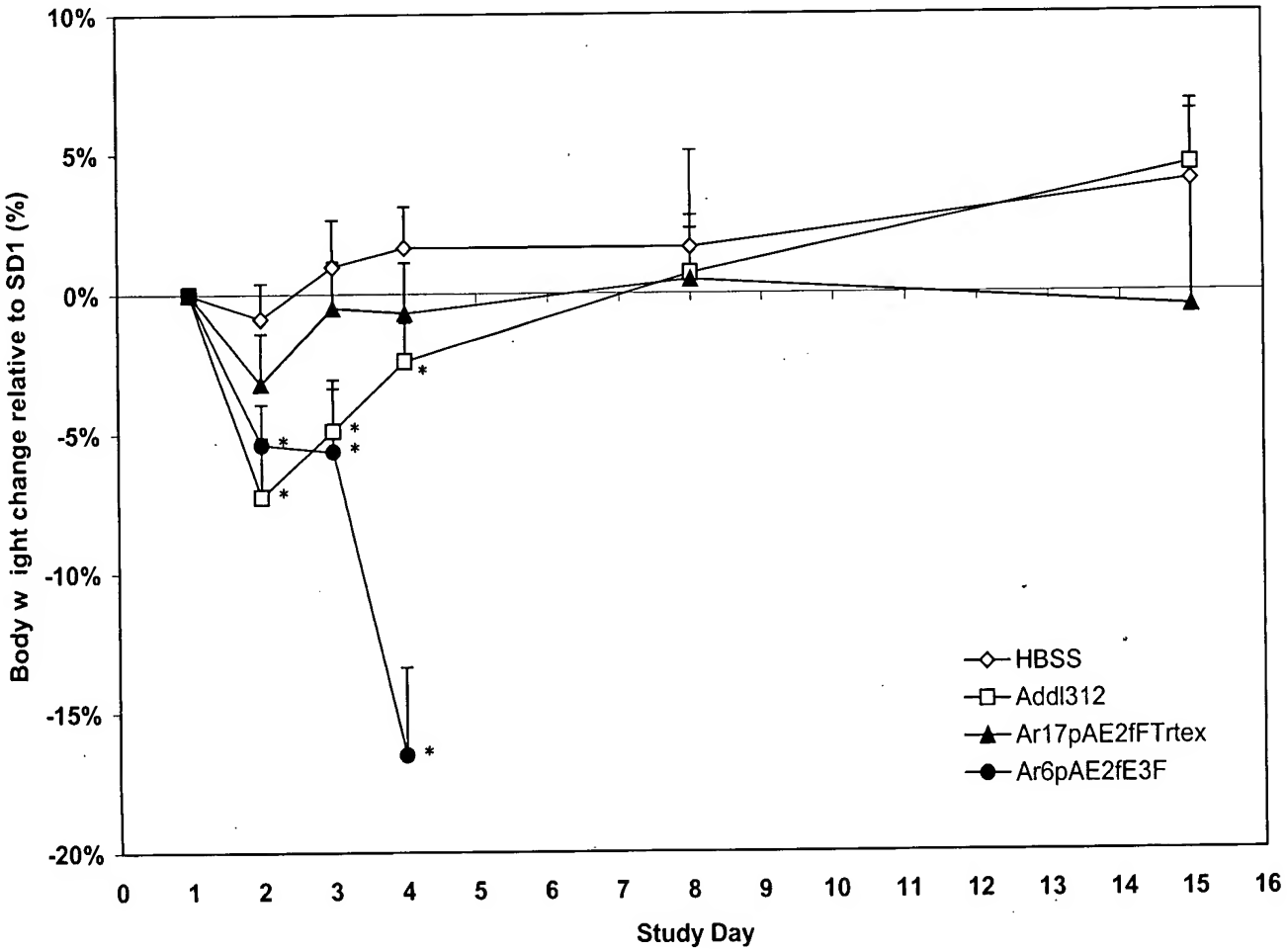
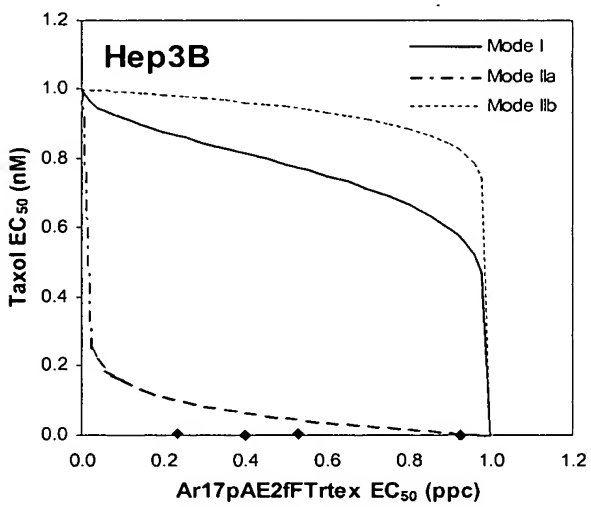
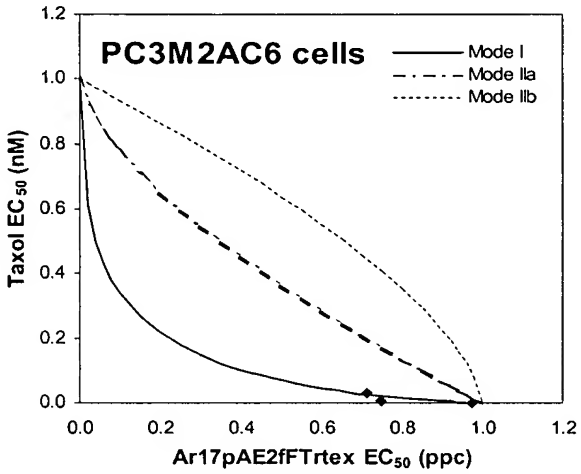


Figure 63. Improved isobologram with additivity envelope for Ar17pAE2fFTrtex and Taxol against Hep 3B and PC3M.2AC6 cells.

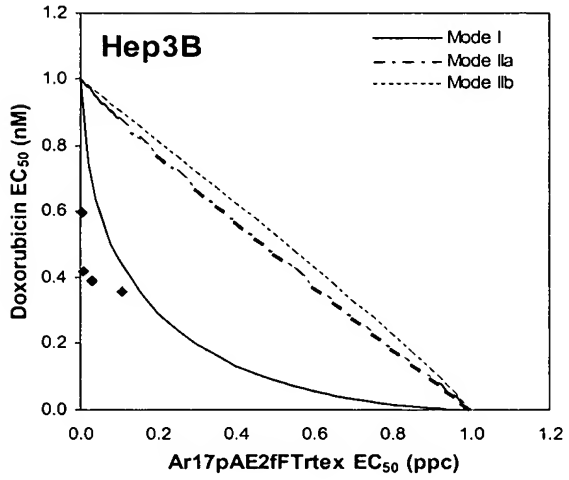


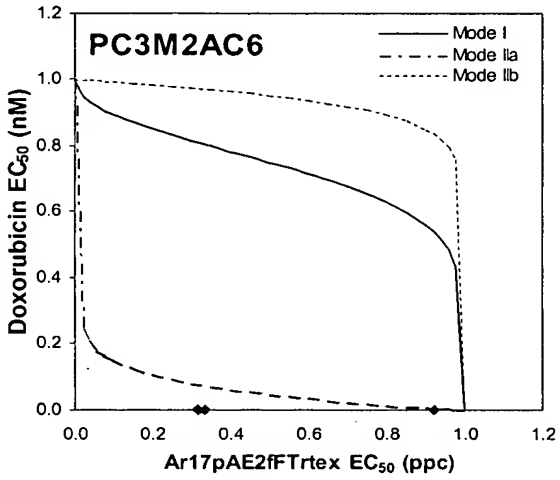


MR (ppc/nM)	Virus EC_{50}^{b}	Chemo EC_{50}^{b}	Effect
Virus alone	1	0	-
Chemo alone	0	1	-
8.3e-05	0.23	0.0043	synergy
3.3e-04	0.53	0.0024	synergy
1.3e-03	0.40	0.00046	synergy
5.3e-03	0.93	0.00027	synergy

MR (ppc/nM)	Virus EC_{50}^{b}	Chemo EC_{50}^{b}	Effect
Virus alone	1	0	-
Chemo alone	0	1	-
0.02	3.4	1.3	antagonism
0.2	0.71	0.028	synergy
2	0.75	0.003	synergy
20	0.97	0.0004	synergy

Figure 64. Improved isobologram with additivity envelope for Ar17pAE2fFTrtex and Doxorubicin against Hep 3B and PC3M.2AC6 cells.

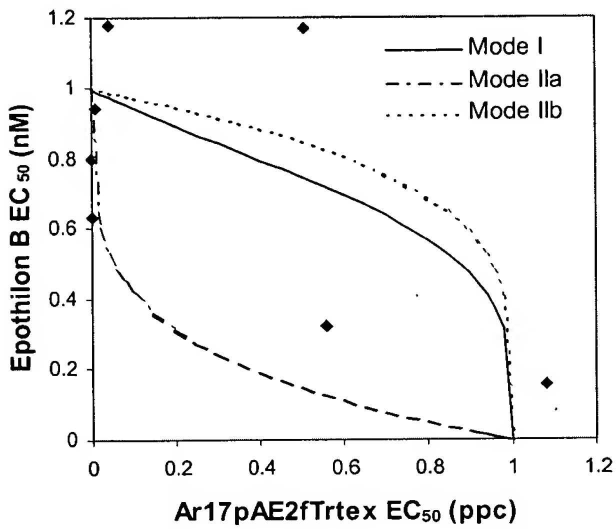




MR (ppc/nM)	Virus EC_{50}^{b}	Chemo EC_{50}^{b}	Effect
Virus alone	1	0	-
Chemo alone	0	1	-
1.3e-05	0.0028	0.60	synergy
5.0e-05	0.0078	0.42	synergy
2.0e-04	0.029	0.39	synergy
8.0e-04	0.11	0.36	synergy

MR (ppc/nM)	Virus EC_{50}^{b}	Chemo EC_{50}^{b}	Effect
Virus alone	1	0	-
Chemo alone	0	1	-
1	2.2	0.015	antagonism
10	0.92	6.1e-4	synergy
100	0.34	2.2e-5	synergy
1000	0.32	2.1e-6	synergy

Figure 65. Improved isobologram with additivity envelope for Ar17pAE2fFTrtex and Epothilone B against Hep 3B cells.



	Virus EC_{50}^{b}	Chemo EC_{50}^{b}	Effect
Virus alone	1	0	-
Chemo alone	0	1	-
3.1e-06	0.00045	0.63	synergy
1.3e-05	0.0018	0.80	synergy
5.0e-05	0.0084	0.95	synergy
2.0e-04	0.042	1.2	antagonism
8.0e-04	0.18	1.6	antagonism
3.2e-03	0.51	1.2	antagonism
1.3e-02	0.56	0.32	additivity
5.1e-02	1.1	0.06	antagonism

Figure 66. Doxorubicin Combination: Mean Tumor Volumes

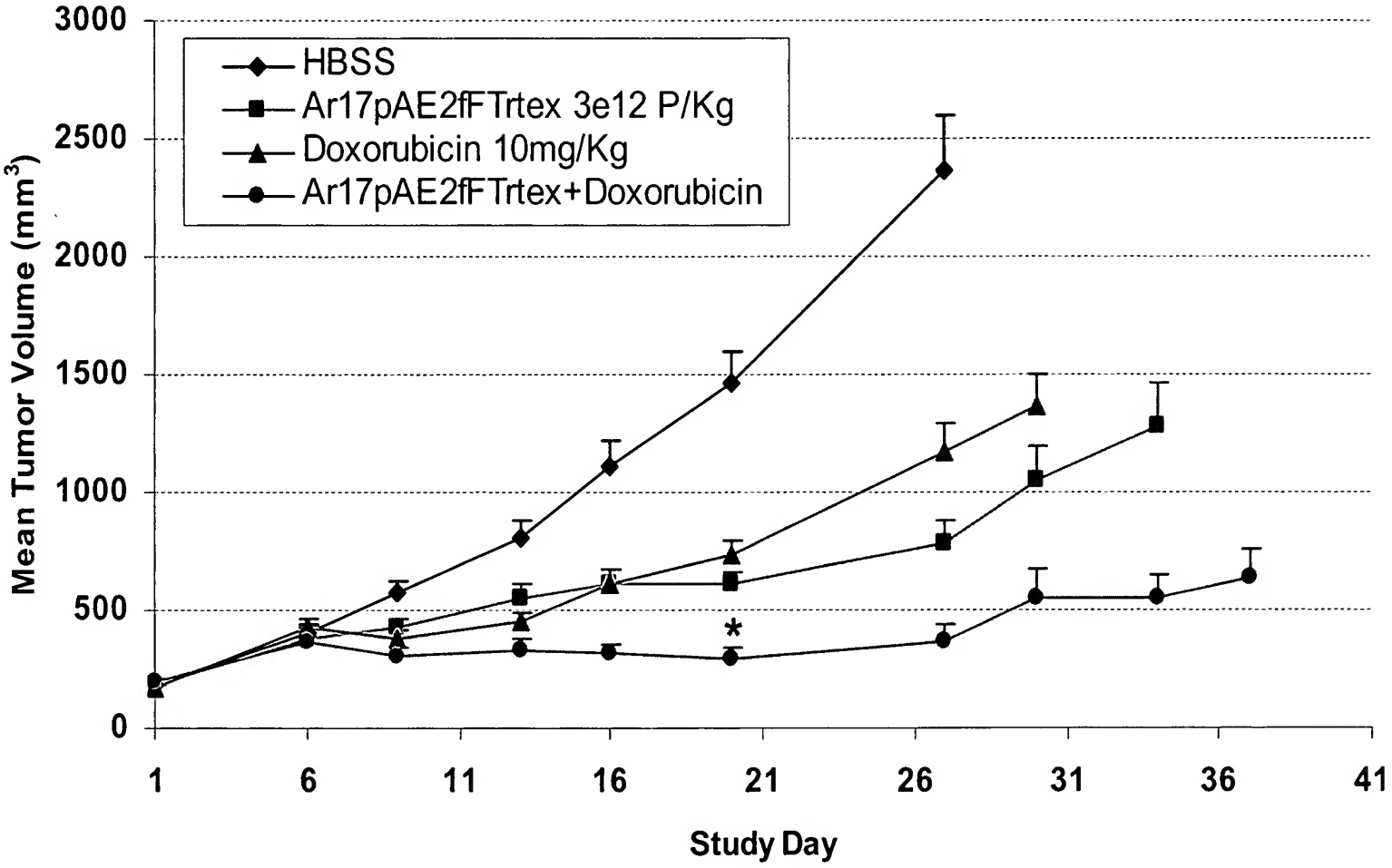


Figure 67. Doxil® Combination Mean Tumor Volumes

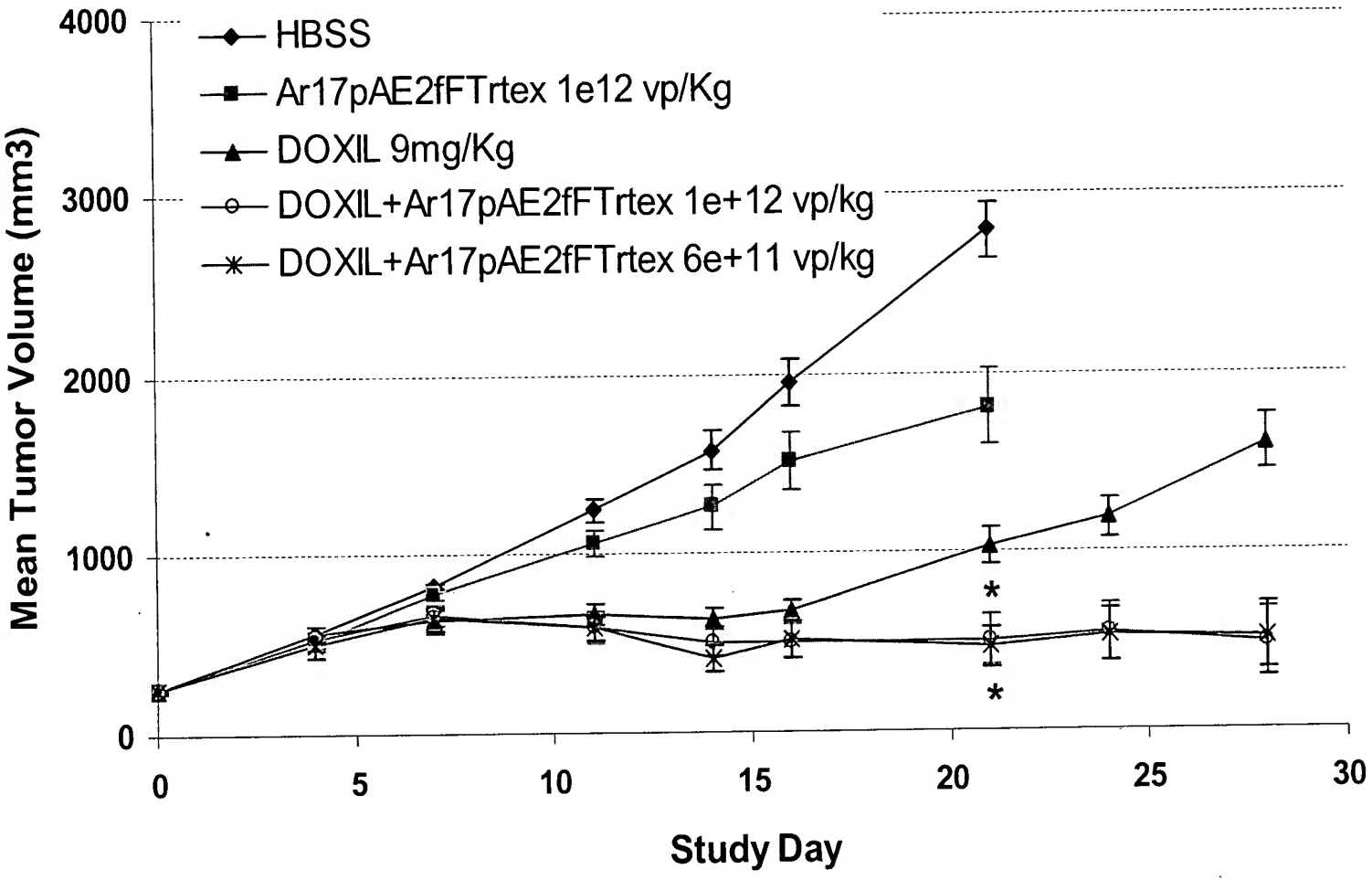


Figure 68. Cytotoxicity assessed in primary human hepatocytes

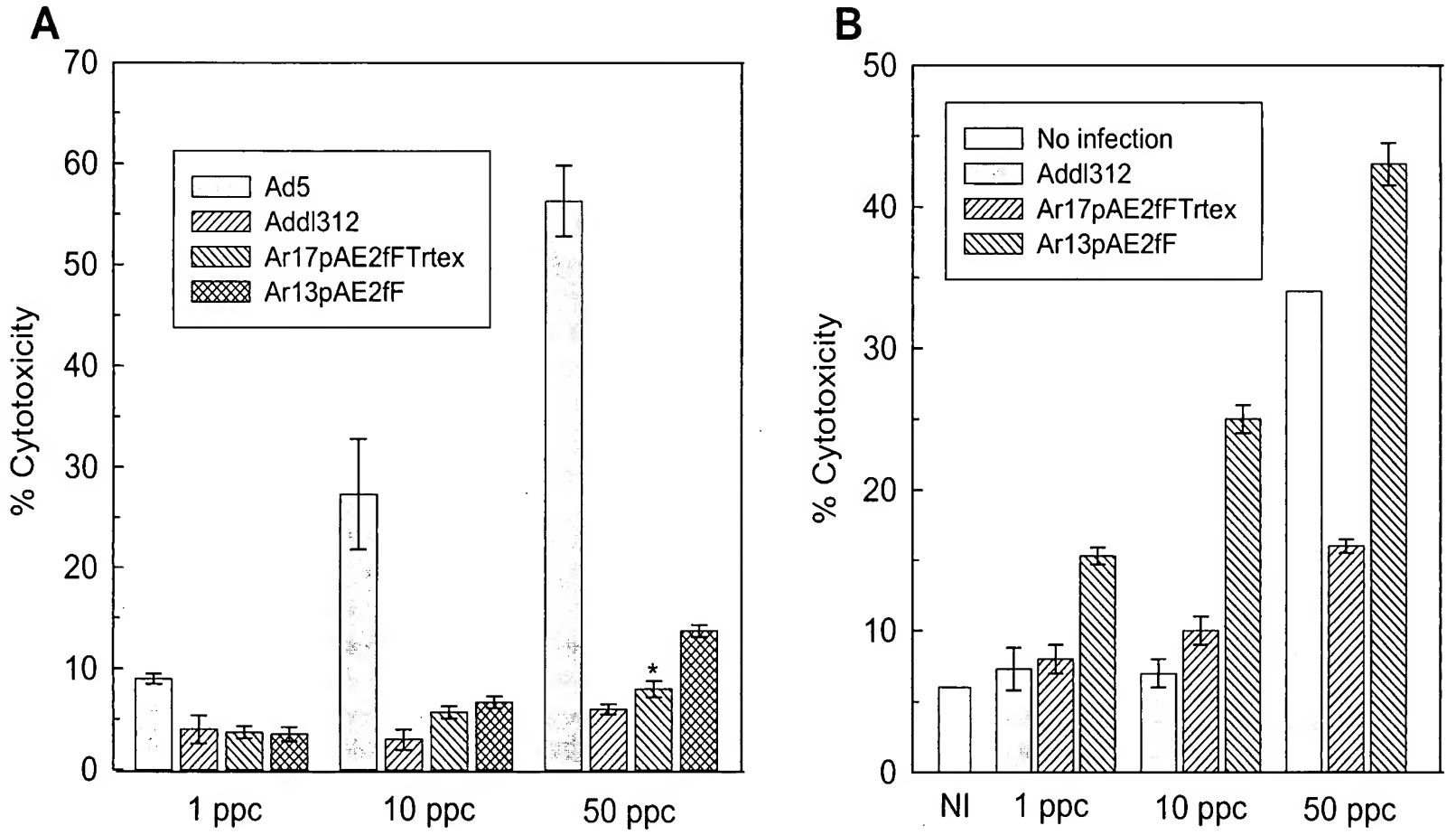


Figure 69

Ad35-Based Oncolytic Vectors

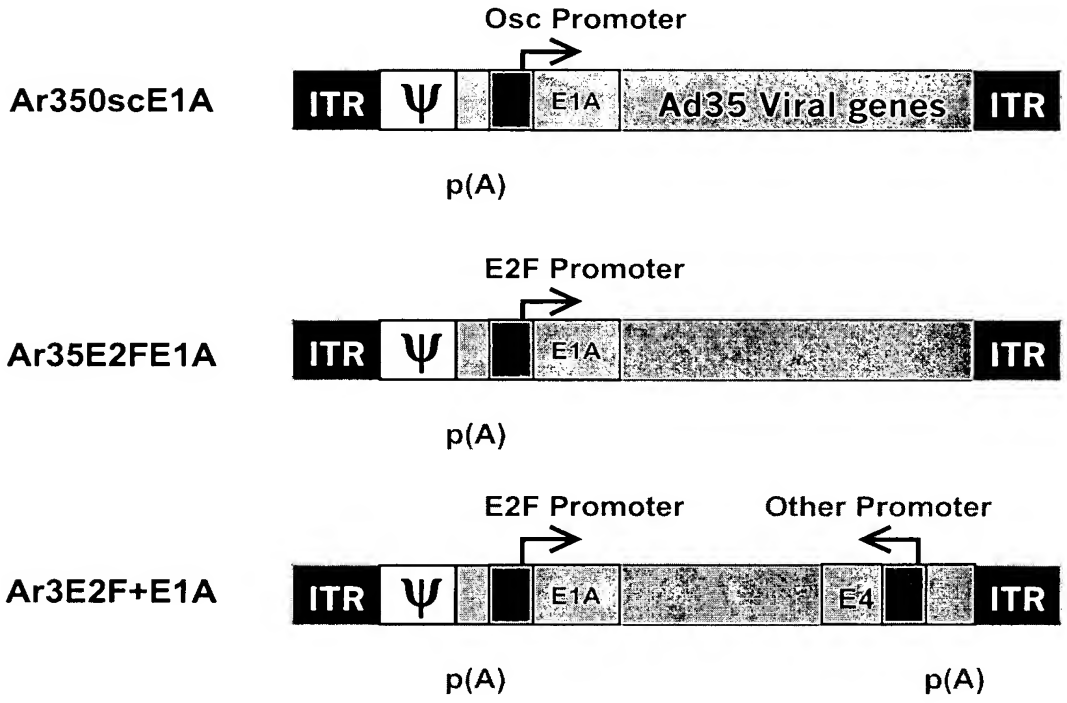
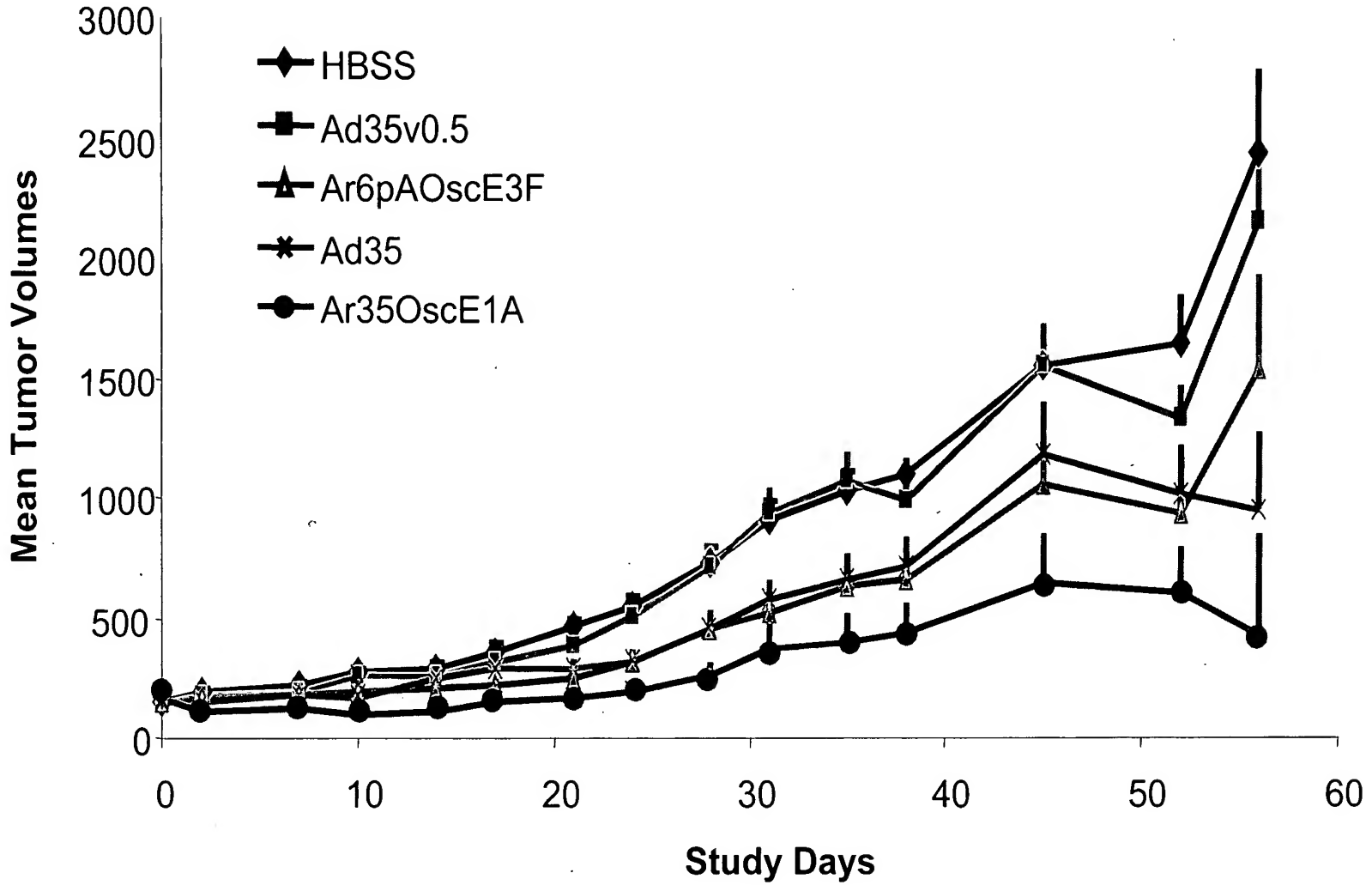


Figure 70


HBSS
Ad35v0.5
Ar6pAOscE3F
Ad35
Ar35OscE1A
Mean Tumor Volumes
3000
2500
2000
1500
1000
500
0
0
10
20
30
40
50
60
Study Days